Fig. 1.a: Construction of a vector for the expression of an rML-ITF of the TPE type (bFGF-MLA)

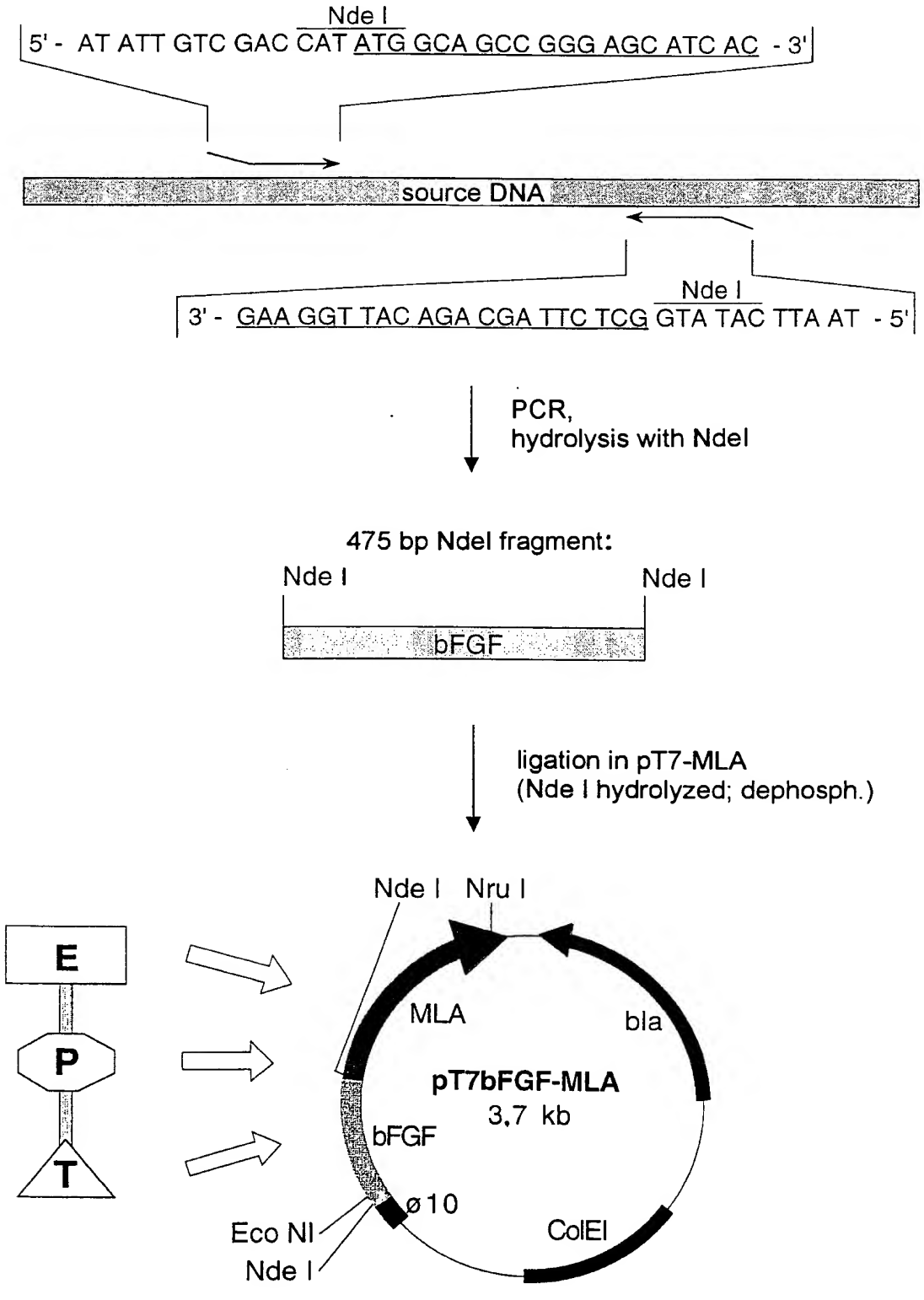


Fig. 1.b: C-terminal processing sequence of bFGF

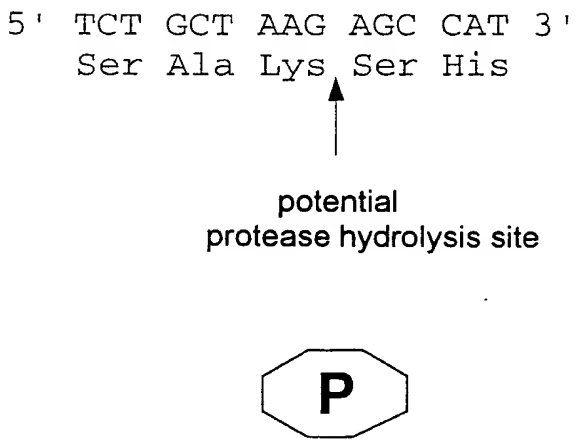


Fig. 1.c: Expression vector of the effector module (rMLA)

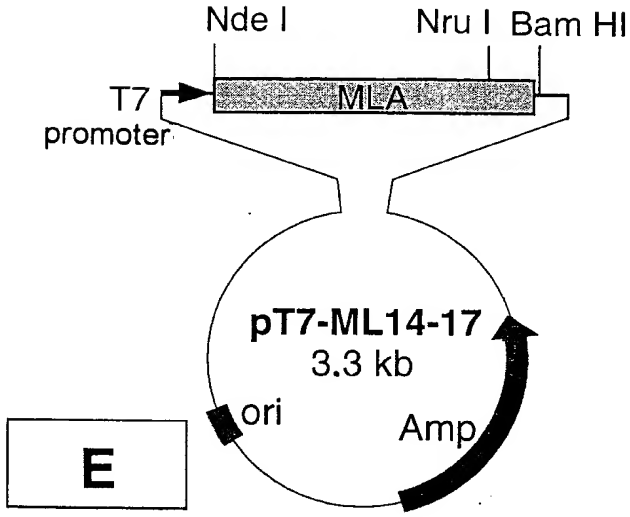
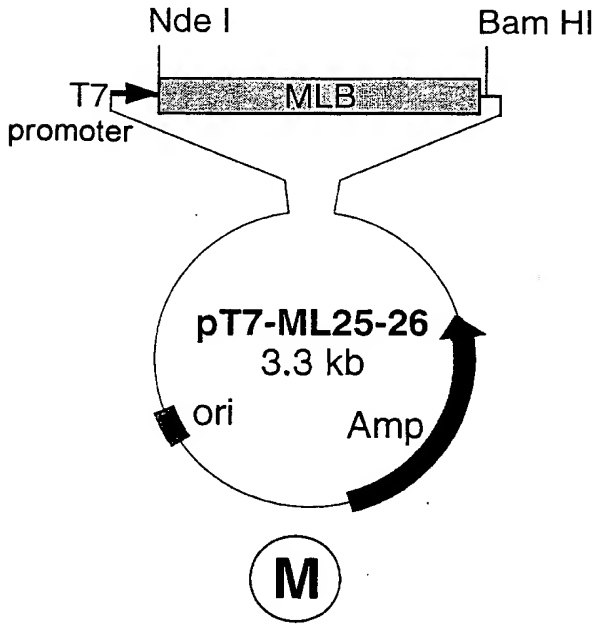


Fig. 2: Vectors for the expression of the modules TPE (bFGF-MLA) and M (rMLB) for in vitro association



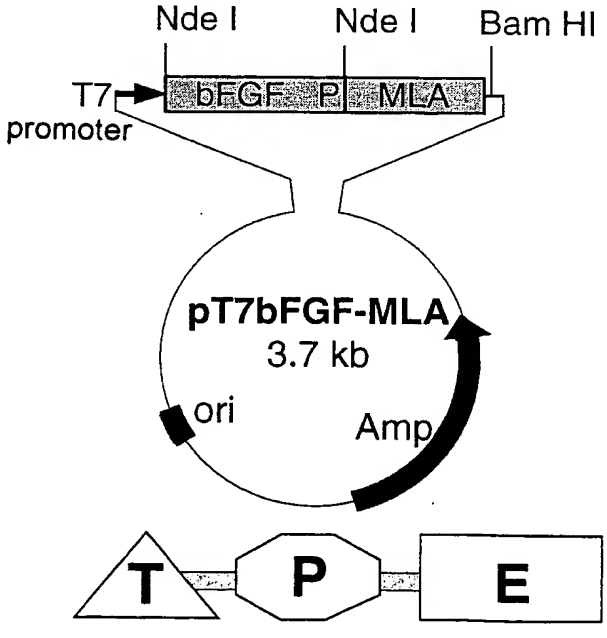


Fig. 3: Construction of a vector for the expression of an ML-ITF of the EPM^T type (ProML)

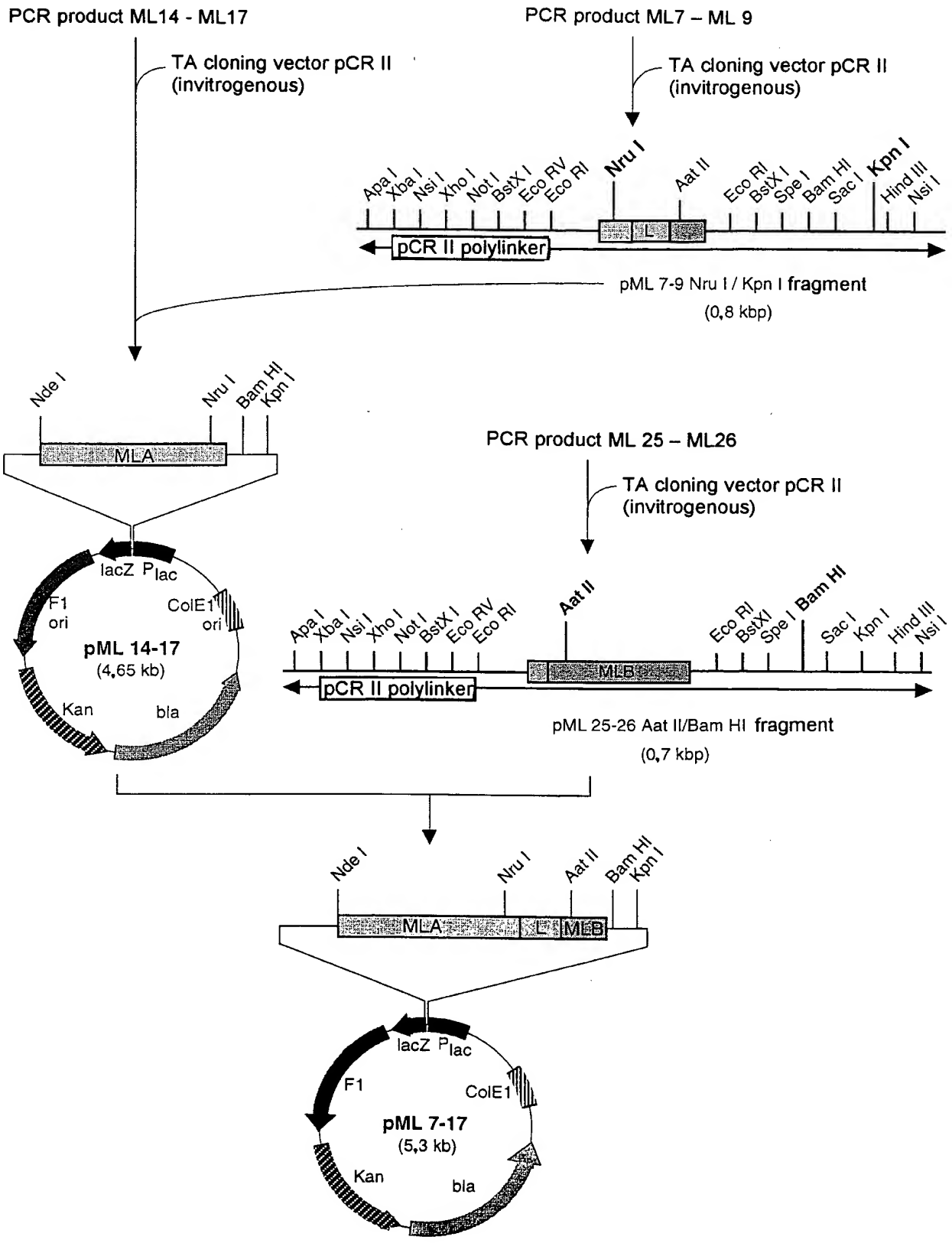


Fig. 3 continued:

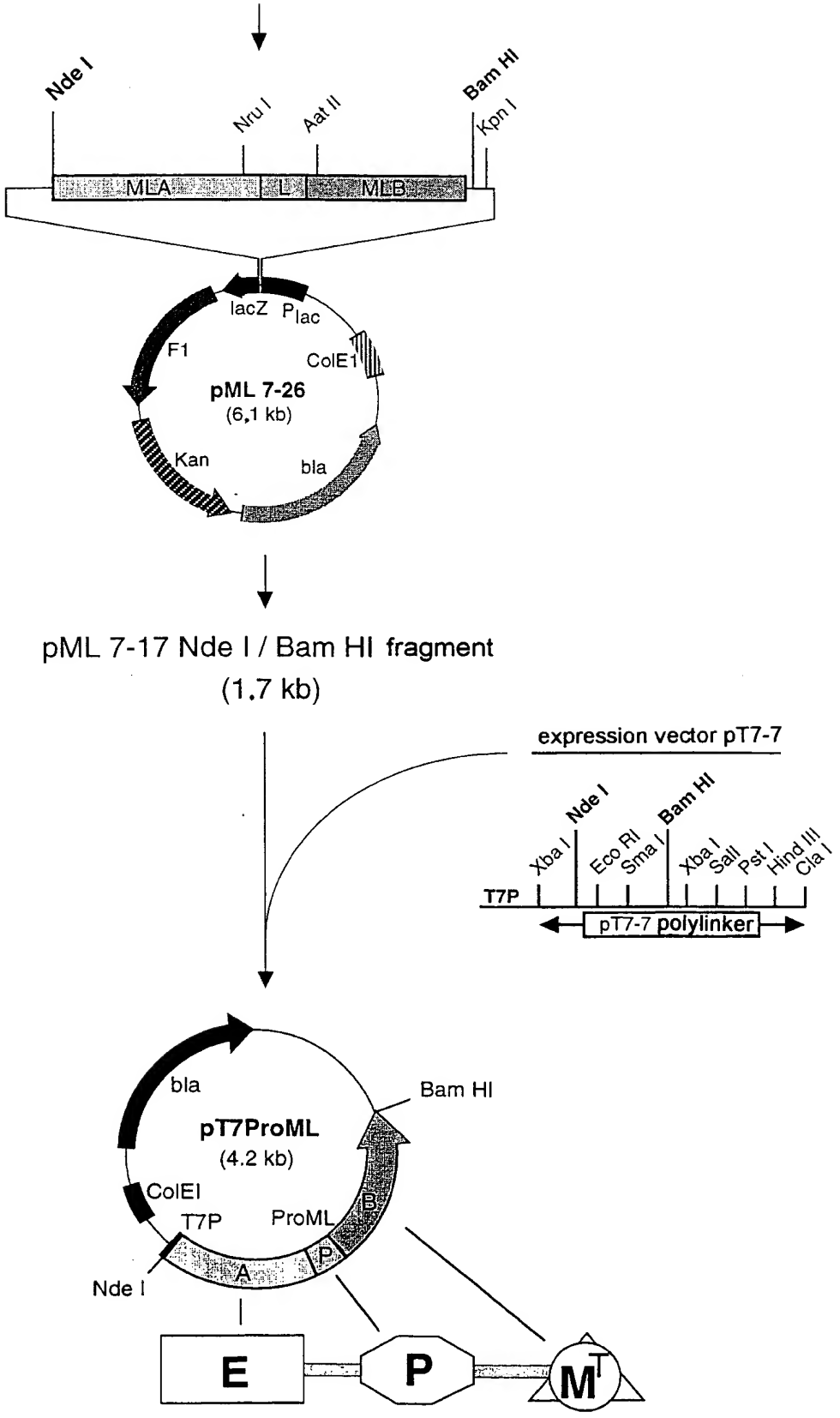


Fig. 4.a: Recombinant production of bFGF-MLA

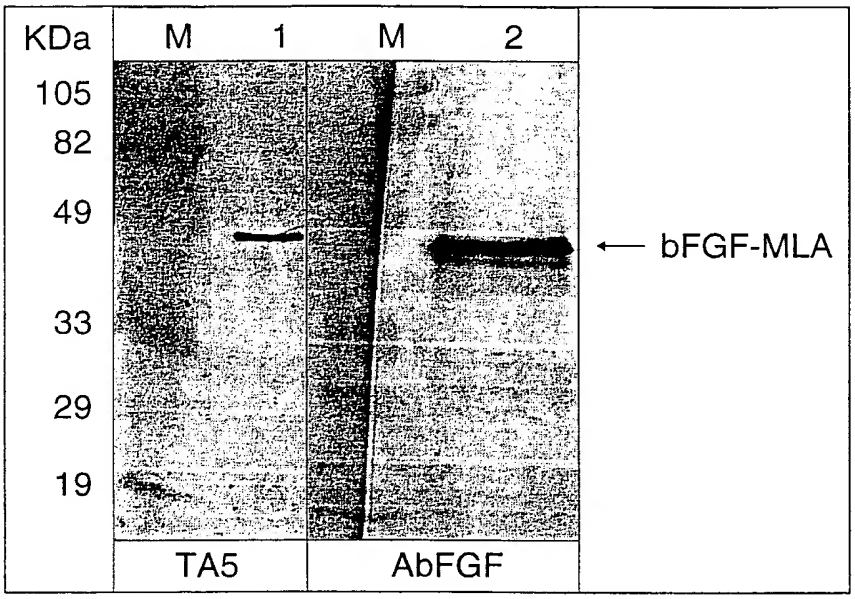


Fig. 4.b: Recombinant production of rMLA

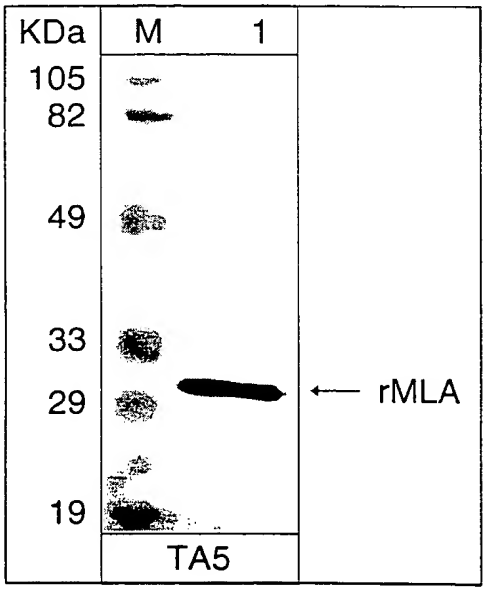


Fig. 5.a: Recombinant production of bFGF-MLA/rMLB (total protein stain)

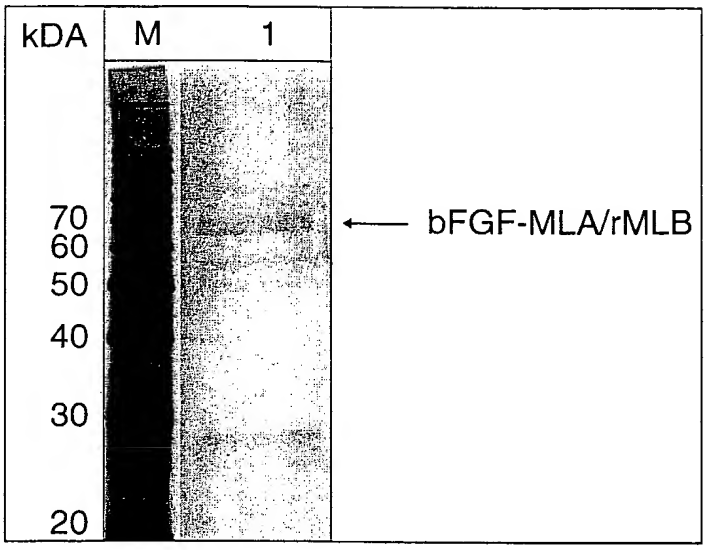


Fig. 5.b: Recombinant production of bFGF-MLA/rMLB (Western blot analysis)

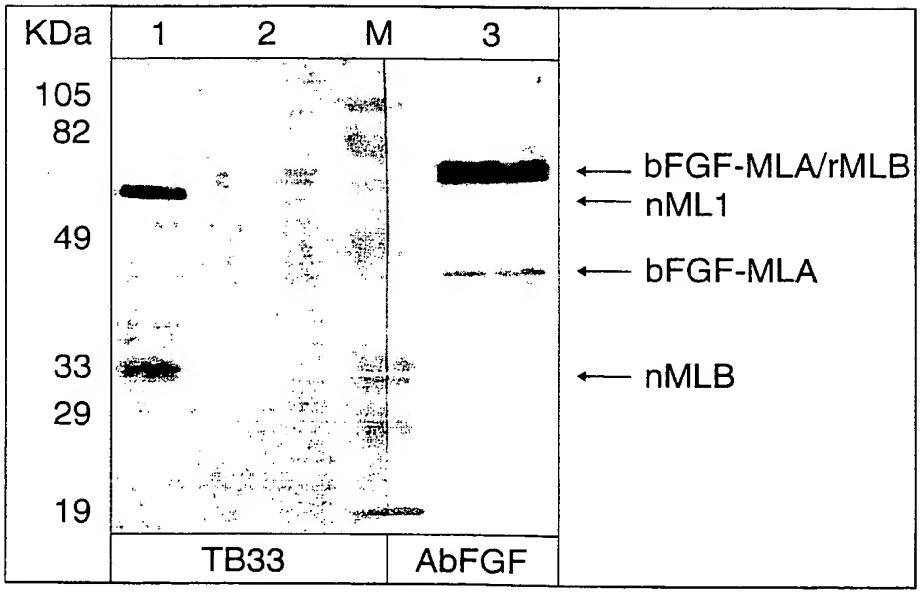


Fig. 6: Recombinant production of ProML

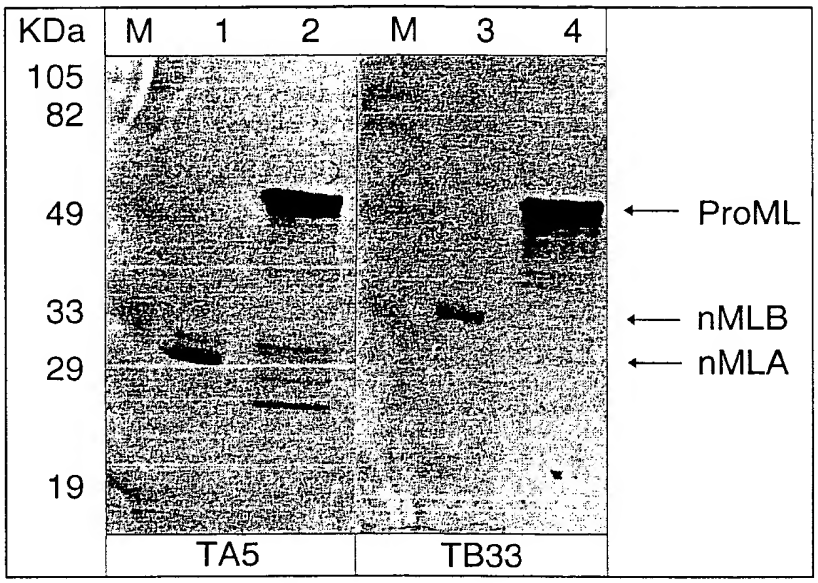


Fig. 7: Cytotoxicity of bFGF-MLA

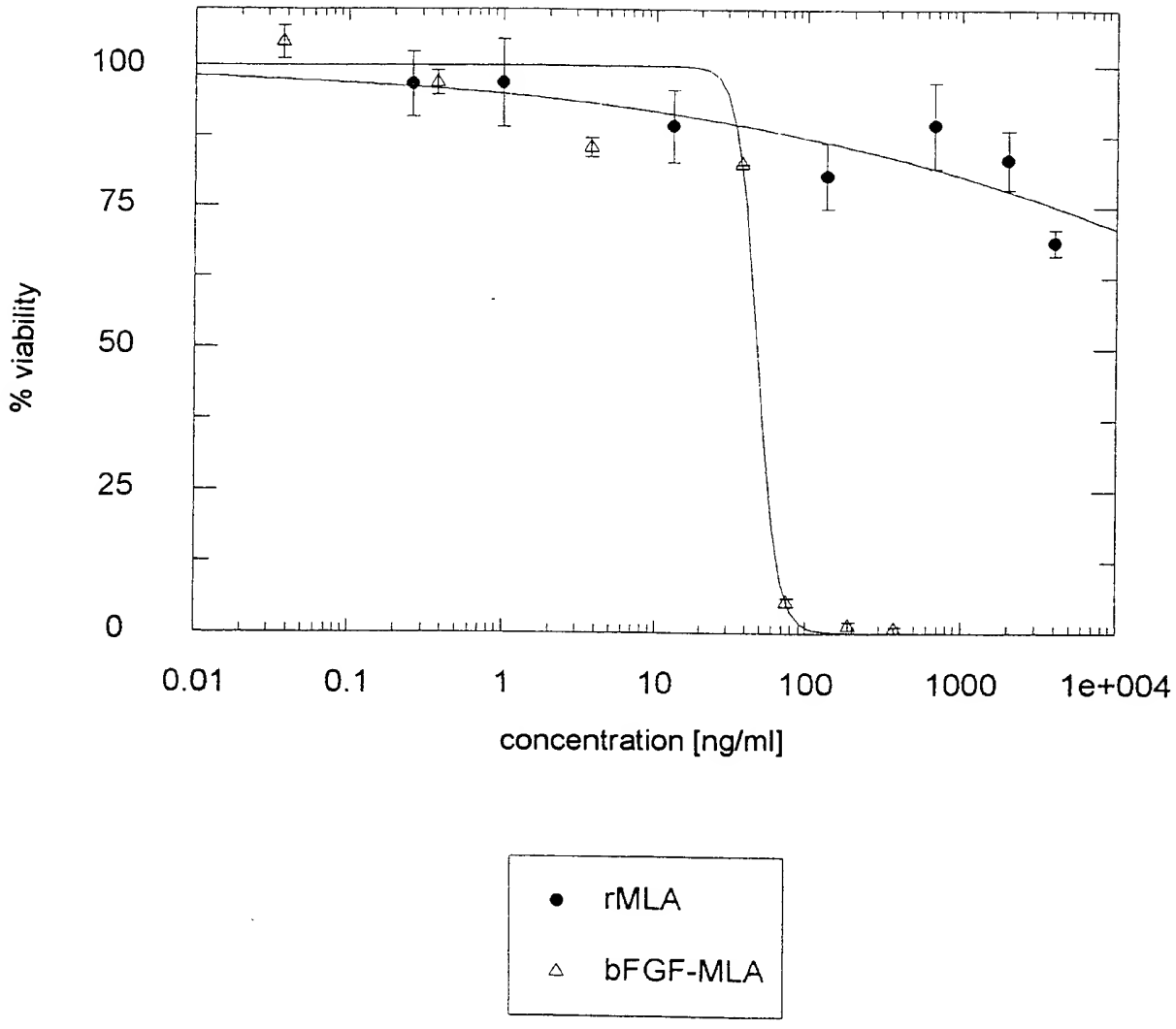


Fig. 8.a: Cytotoxicity of bFGF-MLA/rMLB

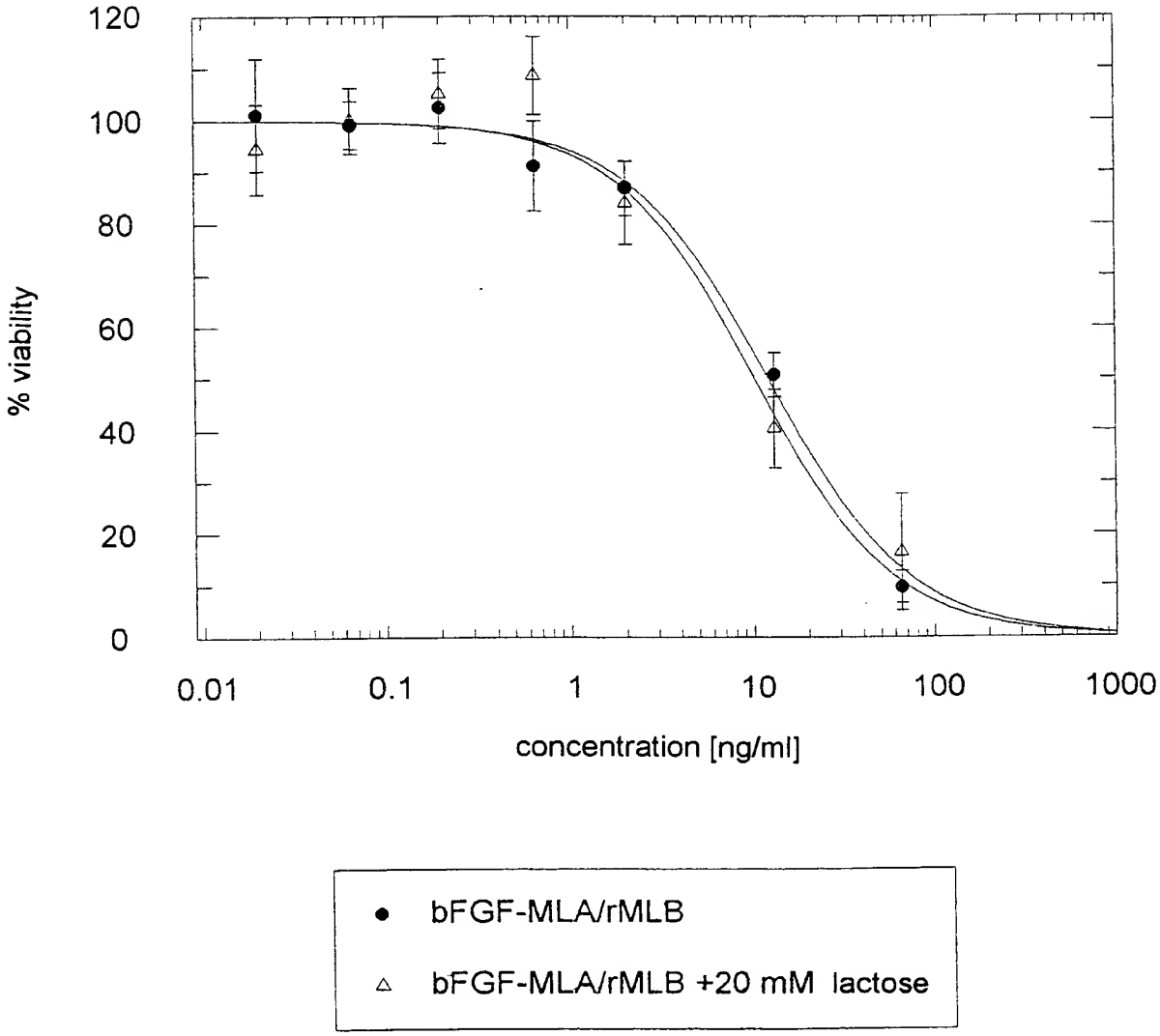


Fig. 8.b: Modulation of the cytotoxicity of bFG-MLA by rMLB

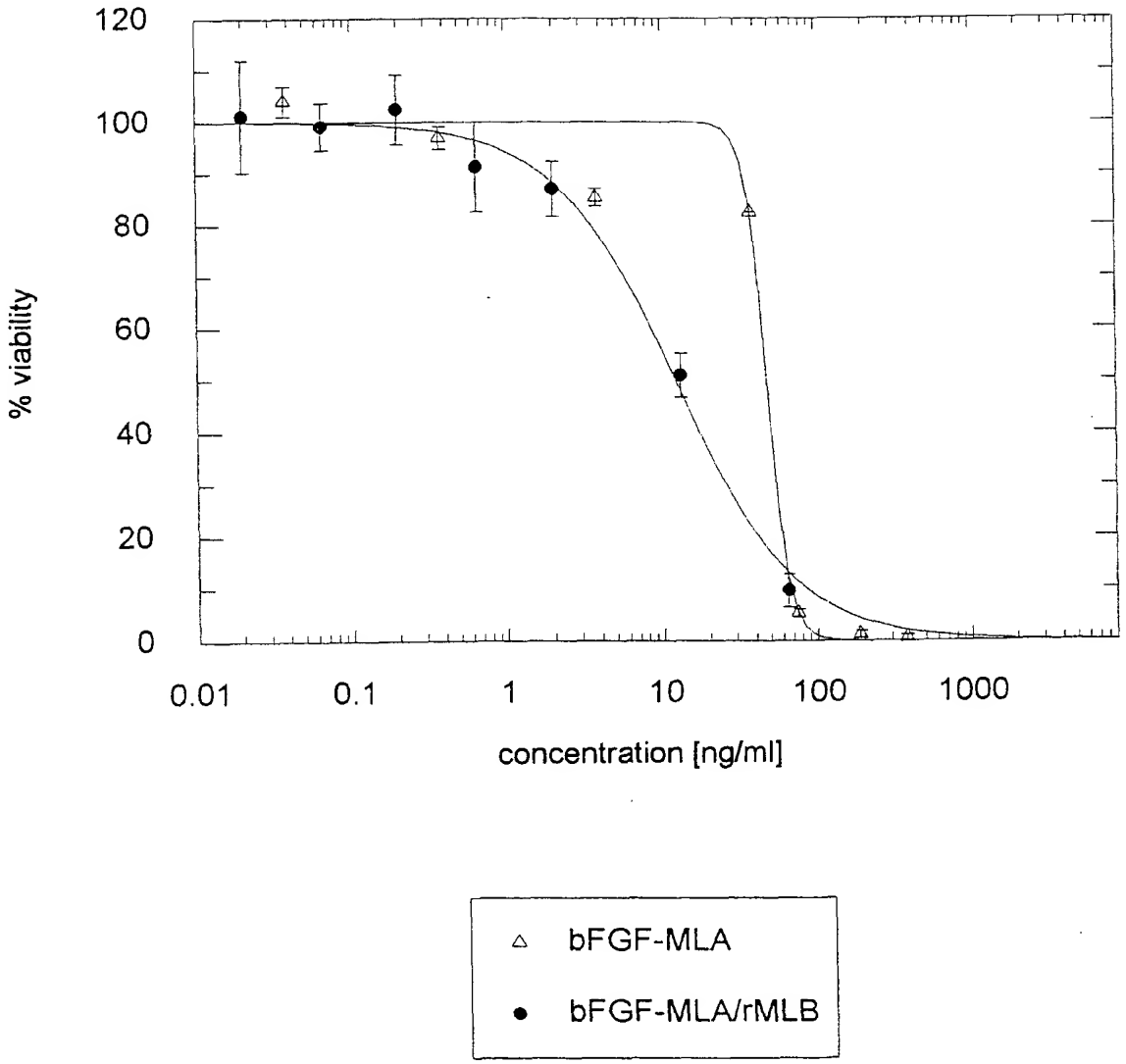


Fig. 9.a: Cytotoxicity of ProML

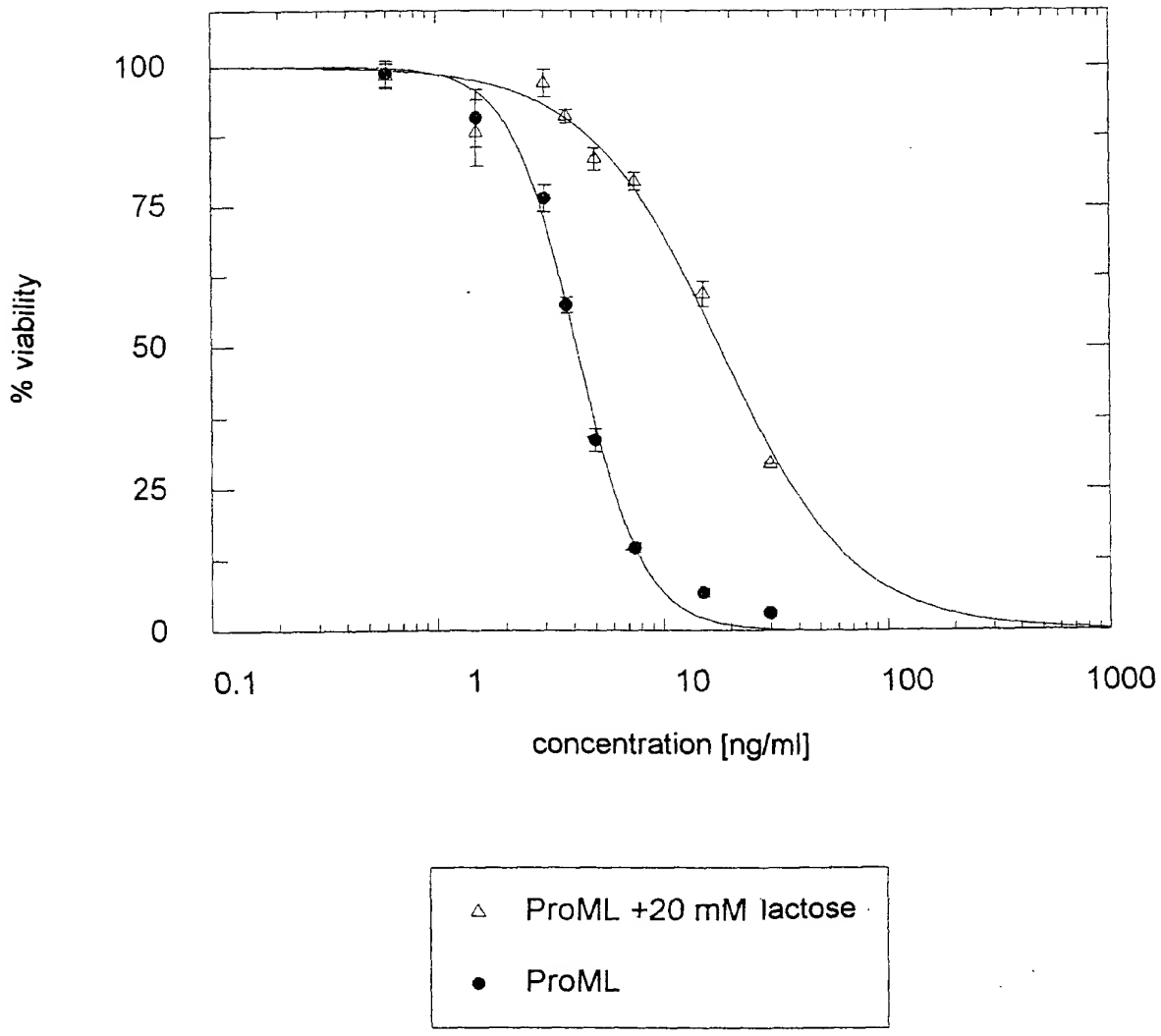


Fig. 9.b: Cytotoxicity of ProML as compared to rML

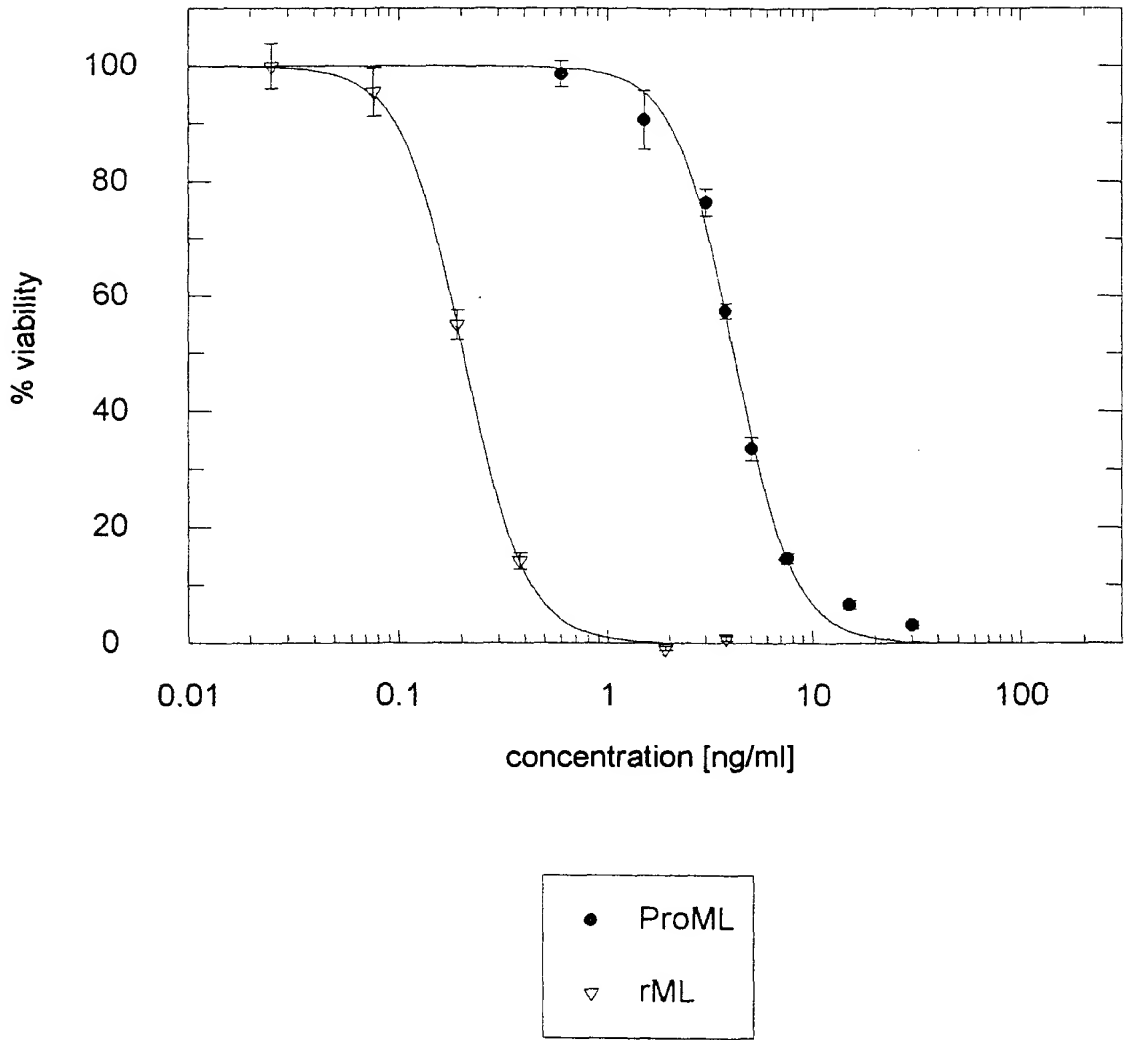
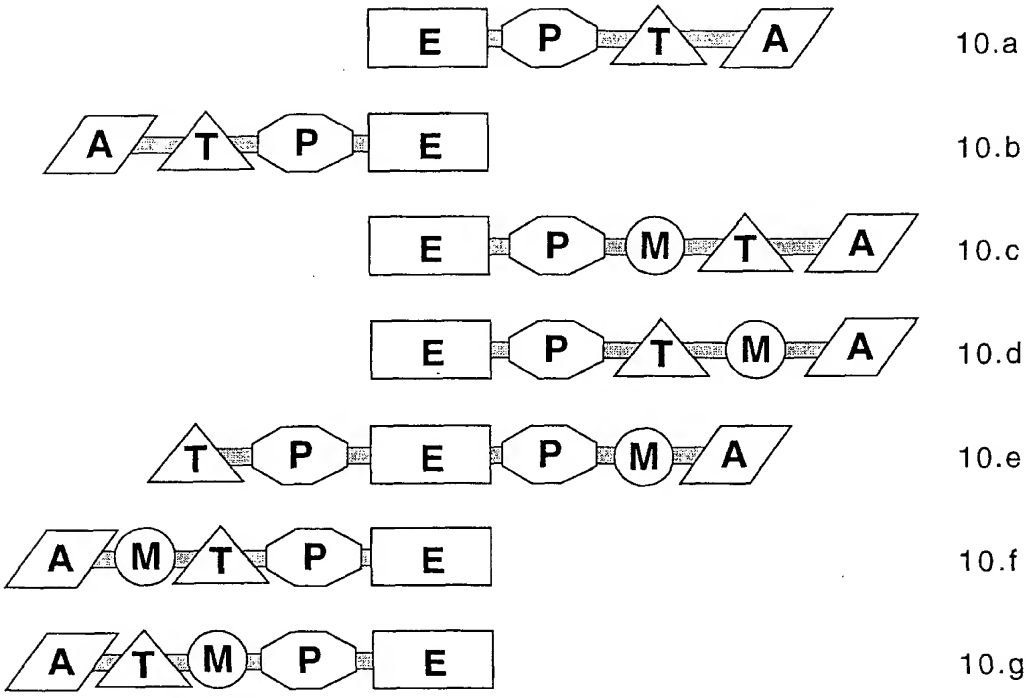


Fig. 10: Exemplary selection of possible combinations of the rML-ITF modules



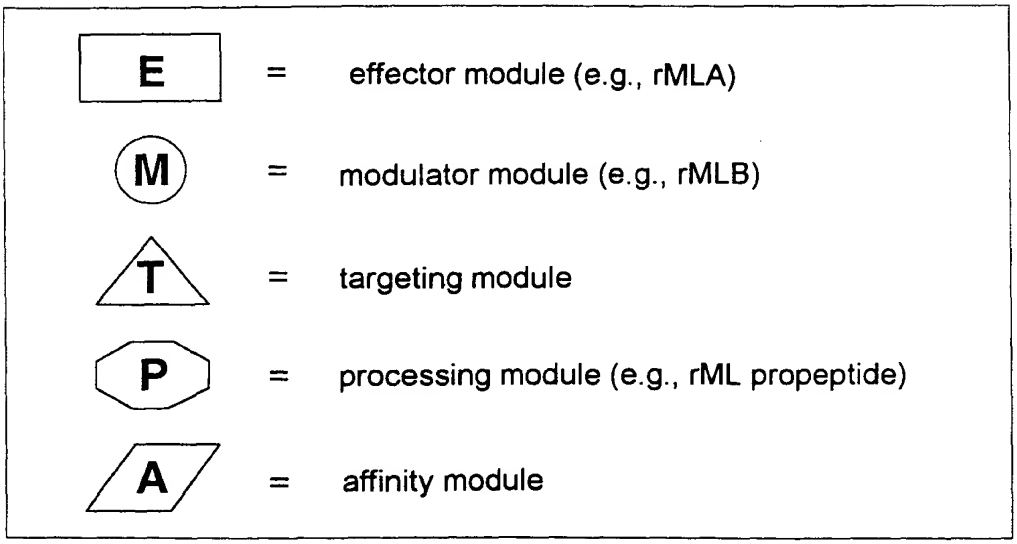


Fig. 11.a: Nucleotide sequence and derived amino acid sequence of rMLA

```
    NdeI                  18          27          36          45
5'  CAT ATG TAC GAA CGT ATC CGT CTG CGT GTT ACC CAC CAG ACC ACC
        Met Tyr Glu Arg Ile Arg Leu Arg Val Thr His Gln Thr Thr

            54          63          72          81          90
    GGT GAA GAA TAT TTC CGG TTC ATC ACG CTT CTC CGA GAT TAT GTC
    Gly Glu Glu Tyr Phe Arg Phe Ile Thr Leu Leu Arg Asp Tyr Val

            99         108         117         126         135
    TCA AGC GGA AGC TTT TCC AAT GAG ATA CCA CTC TTG CGT CAG TCT
    Ser Ser Gly Ser Phe Ser Asn Glu Ile Pro Leu Leu Arg Gln Ser

           144         153         162         171         180
    ACG ATC CCC GTC TCC GAT GCG CAA AGA TTT GTC TTG GTG GAG CTC
    Thr Ile Pro Val Ser Asp Ala Gln Arg Phe Val Leu Val Glu Leu

           189         198         207         216         225
    ACC AAC CAG GGG GGA GAC TCG ATC ACG GCC GCC ATC GAC GTT ACC
    Thr Asn Gln Gly Gly Asp Ser Ile Thr Ala Ala Ile Asp Val Thr

           234         243         252         261         270
    AAT CTG TAC GTC GTG GCT TAC CAA GCA GGC GAC CAA TCC TAC TTT
    Asn Leu Tyr Val Val Ala Tyr Gln Ala Gly Asp Gln Ser Tyr Phe

           279         288         297         306         315
    TTG CGC GAC GCA CCA CGC GGC GCG GAA ACG CAT CTC TTC ACC GGC
    Leu Arg Asp Ala Pro Arg Gly Ala Glu Thr His Leu Phe Thr Gly

           324         333         342         351         360
    ACC ACC CGA TCC TCT CTC CCA TTC AAC GGA AGC TAC CCT GAT CTG
    Thr Thr Arg Ser Ser Leu Pro Phe Asn Gly Ser Tyr Pro Asp Leu

           369         378         387         396         405
    GAG CGA TAC GCC GGA CAT AGG GAC CAG ATC CCT CTC GGT ATA GAC
    Glu Arg Tyr Ala Gly His Arg Asp Gln Ile Pro Leu Gly Ile Asp

           414         423         432         441         450
    CAA CTC ATT CAA TCC GTC ACG GCG CTT CGT TTT CCG GGC GGC AGC
    Gln Leu Ile Gln Ser Val Thr Ala Leu Arg Phe Pro Gly Gly Ser

           459         468         477         486         495
    ACG CGT ACC CAA GCT CGT TCG ATT TTA ATC CTC ATT CAG ATG ATC
    Thr Arg Thr Gln Ala Arg Ser Ile Leu Ile Leu Ile Gln Met Ile

           504         513         522         531         540
    TCC GAG GCC GCC AGA TTC AAT CCC ATC TTA TGG AGG GCT CGC CAA
    Ser Glu Ala Ala Arg Phe Asn Pro Ile Leu Trp Arg Ala Arg Gln
```

Fig. 11.a continued:

```
          549          558          567          576          585
TAC ATT AAC AGT GGG GCG TCA TTT CTG CCA GAC GTG TAC ATG CTG
Tyr Ile Asn Ser Gly Ala Ser Phe Leu Pro Asp Val Tyr Met Leu

          594          603          612          621          630
GAG CTG GAG ACG AGT TGG GGC CAA CAA TCC ACG CAA GTC CAG CAT
Glu Leu Glu Thr Ser Trp Gly Gln Gln Ser Thr Gln Val Gln His

          639          648          657          666          675
TCA ACC GAT GGC GTT TTT AAT AAC CCA ATT CGG TTG GCT ATA CCC
Ser Thr Asp Gly Val Phe Asn Asn Pro Ile Arg Leu Ala Ile Pro

          684          693          702          729          738
CCC GGT AAC TTC GTG ACG TTG ACC AAT GTT CGC GAC GTG ATC GCC
Pro Gly Asn Phe Val Thr Leu Thr Asn Val Arg Asp Val Ile Ala

          747          756          765            AvaI
AGC TTG GCG ATC ATG TTG TTT GTA TGC GGA GAG CGC CCG AGT  3'
Ser Leu Ala Ile Met Leu Phe Val Cys Gly Glu Arg Pro
```

Fig. 11.b: Nucleotide sequence and derived amino acid sequence of rMLB

```
    StuI      9           18          BspLU11I     36          45
5' AGG CCT GTG ATA GCC GAT GAT GTT ACA TGT AGT GCT TCG GAA CCT
                        Asp Asp Val Thr Cys Ser Ala Ser Glu Pro

           54          63          72          81          90
ACG GTG CGG ATT GTG GGT CGA AAT GGC ATG TGC GTG GAC GTC CGA
Thr Val Arg Ile Val Gly Arg Asn Gly Met Cys Val Asp Val Arg

           99          108         117         126         135
GAT GAC GAT TTC CGC GAT GGA AAT CAG ATA CAG TTG TGG CCC TCC
Asp Asp Asp Phe Arg Asp Gly Asn Gln Ile Gln Leu Trp Pro Ser

          144          153         162         171         180
AAG TCC AAC AAT GAT CCG AAT CAG TTG TGG ACG ATC AAA AGG GAT
Lys Ser Asn Asn Asp Pro Asn Gln Leu Trp Thr Ile Lys Arg Asp

          189          198         207         216         225
GGA ACC ATT CGA TCC AAT GGC AGC TGC TTG ACC ACG TAT GGC TAT
Gly Thr Ile Arg Ser Asn Gly Ser Cys Leu Thr Thr Tyr Gly Tyr
```

Fig. 11.b continued:

```
        234         243         252         261         270
ACT GCT GGC GTC TAT GTG ATG ATC TTC GAC TGT AAT ACT GCT GTG
Thr Ala Gly Val Tyr Val Met Ile Phe Asp Cys Asn Thr Ala Val

        279         288         297         306         315
CGG GAG GCC ACT CTT TGG CAG ATA TGG GGC AAT GGG ACC ATC ATC
Arg Glu Ala Thr Leu Trp Gln Ile Trp Gly Asn Gly Thr Ile Ile

        324         333         342         351         360
AAT CCA AGA TCC AAT CTG GTT TTG GCA GCA TCA TCT GGA ATC AAA
Asn Pro Arg Ser Asn Leu Val Leu Ala Ala Ser Ser Gly Ile Lys

        369         378         387         396         405
GGC ACT ACG CTT ACG GTG CAA ACA CTG GAT TAC ACG TTG GGA CAG
Gly Thr Thr Leu Thr Val Gln Thr Leu Asp Tyr Thr Leu Gly Gln

        414         423         432         441         450
GGC TGG CTT GCC GGT AAT GAT ACC GCC CCA CGC GAG GTG ACC ATA
Gly Trp Leu Ala Gly Asn Asp Thr Ala Pro Arg Glu Val Thr Ile

        459         468         477         486         495
TAT GGG TTC AGG GAC CTT TGC ATG GAA TCA AAT GGA GGG AGT GTG
Tyr Gly Phe Arg Asp Leu Cys Met Glu Ser Asn Gly Gly Ser Val

        504         513         522         531         540
TGG GTG GAG ACG TGC GTG AGT AGC CAA AAG AAC CAA AGA TGG GCT
Trp Val Glu Thr Cys Val Ser Ser Gln Lys Asn Gln Arg Trp Ala

        549         558         567         576         585
TTG TAC GGG GAT GGT TCT ATA CGC CCC AAA CAA AAC CAA GAC CAA
Leu Tyr Gly Asp Gly Ser Ile Arg Pro Lys Gln Asn Gln Asp Gln

        594         603         612         621         630
TGC CTC ACC TGT GGG AGA GAC TCC GTT TCA ACA GTA ATC AAT ATA
Cys Leu Thr Cys Gly Arg Asp Ser Val Ser Thr Val Ile Asn Ile

        639         648         657         666         675
GTT AGC TGC AGC GCT GGA TCG TCT GGG CAG CGA TGG GTG TTT ACC
Val Ser Cys Ser Ala Gly Ser Ser Gly Gln Arg Trp Val Phe Thr

        684         693         702         711         720
AAT GAA GGG GCC ATT TTG AAT TTA AAG AAT GGG TTG GCC ATG GAT
Asn Glu Gly Ala Ile Leu Asn Leu Lys Asn Gly Leu Ala Met Asp

        729         738         747         756         765
GTG GCG CAA GCA AAT CCA AAG CTC CGC CGA ATA ATC ATC TAT CCT
Val Ala Gln Ala Asn Pro Lys Leu Arg Arg Ile Ile Ile Tyr Pro

        774         783         792         801          ___
GCC ACA GGA AAA CCA AAT CAA ATG TGG CTT CCC GTG CCA GGT GGA
Ala Thr Gly Lys Pro Asn Gln Met Trp Leu Pro Val Pro Gly Gly

EcoRV   819      BamHI
TAT CAC TAG TAA GGA TCC  3'
Tyr His *** ***
```

Fig. 11.c: Nucleotide sequence and derived amino acid sequence of the rML propeptide

```
      AvaI                  18          27          36
5' CGC CCG AGT TCC TCT GAG GTG CGC TAT TGG CCG CTG
          Ser Ser Ser Glu Val Arg Tyr Trp Pro Leu

          StuI                      63      BspLU11I
   GTC ATA AGG CCT GTG ATA GCC GAT GAT GTT ACA TGT 3'
   Val Ile Arg Pro Val Ile Ala
```

Fig. 11.a': Nucleotide sequence and derived amino acid sequence of rMLA

```
                9          18          27          36          45
5'- TAC GAA CGT ATC CGT CTG CGT GTT ACC CAC CAG ACC ACC GGT GAA
    Tyr Glu Arg Ile Arg Leu Arg Val Thr His Gln Thr Thr Gly Glu

               54          63          72          81          90
    GAA TAT TTC CGG TTC ATC ACG CTT CTC CGA GAT TAT GTC TCA AGC
    Glu Tyr Phe Arg Phe Ile Thr Leu Leu Arg Asp Tyr Val Ser Ser

               99         108         117         126         135
    GGA AGC TTT TCC AAT GAG ATA CCA CTC TTG CGT CAG TCT ACG ATC
    Gly Ser Phe Ser Asn Glu Ile Pro Leu Leu Arg Gln Ser Thr Ile

              144         153         162         171         180
    CCC GTC TCC GAT GCG CAA AGA TTT GTC TTG GTG GAG CTC ACC AAC
    Pro Val Ser Asp Ala Gln Arg Phe Val Leu Val Glu Leu Thr Asn

              189         198         207         216         225
    CAG GGG GGA GAC TCG ATC ACG GCC GCC ATC GAC GTT ACC AAT CTG
    Gln Gly Gly Asp Ser Ile Thr Ala Ala Ile Asp Val Thr Asn Leu

              234         243         252         261         270
    TAC GTC GTG GCT TAC CAA GCA GGC GAC CAA TCC TAC TTT TTG CGC
    Tyr Val Val Ala Tyr Gln Ala Gly Asp Gln Ser Tyr Phe Leu Arg

              279         288         297         306         315
    GAC GCA CCA CGC GGC GCG GAA ACG CAT CTC TTC ACC GGC ACC ACC
    Asp Ala Pro Arg Gly Ala Glu Thr His Leu Phe Thr Gly Thr Thr

              324         333         342         351         360
    CGA TCC TCT CTC CCA TTC AAC GGA AGC TAC CCT GAT CTG GAG CGA
    Arg Ser Ser Leu Pro Phe Asn Gly Ser Tyr Pro Asp Leu Glu Arg

               369         378         387         396         405
    TAC GCC GGA CAT AGG GAC CAG ATC CCT CTC GGT ATA GAC CAA CTC
    Tyr Ala Gly His Arg Asp Gln Ile Pro Leu Gly Ile Asp Gln Leu

              414         423         432         441         450
    ATT CAA TCC GTC ACG GCG CTT CGT TTT CCG GGC GGC AGC ACG CGT
    Ile Gln Ser Val Thr Ala Leu Arg Phe Pro Gly Gly Ser Thr Arg

              459         468         477         486         495
    ACC CAA GCT CGT TCG ATT TTA ATC CTC ATT CAG ATG ATC TCC GAG
    Thr Gln Ala Arg Ser Ile Leu Ile Leu Ile Gln Met Ile Ser Glu

              504         513         522         531         540
    GCC GCC AGA TTC AAT CCC ATC TTA TGG AGG GCT CGC CAA TAC ATT
    Ala Ala Arg Phe Asn Pro Ile Leu Trp Arg Ala Arg Gln Tyr Ile
```

Fig. 11.a' continued:

```
          549          558          567          576          585
AAC AGT GGG GCG TCA TTT CTG CCA GAC GTG TAC ATG CTG GAG CTG
Asn Ser Gly Ala Ser Phe Leu Pro Asp Val Tyr Met Leu Glu Leu

          594          603          612          621          630
GAG ACG AGT TGG GGC CAA CAA TCC ACG CAA GTC CAG CAT TCA ACC
Glu Thr Ser Trp Gly Gln Gln Ser Thr Gln Val Gln His Ser Thr

          639          648          657          666          675
GAT GGC GTT TTT AAT AAC CCA ATT CGG TTG GCT ATA CCC CCC GGT
Asp Gly Val Phe Asn Asn Pro Ile Arg Leu Ala Ile Pro Pro Gly

          684          693          702          711          720
AAC TTC GTG ACG TTG ACC AAT GTT CGC GAC GTG ATC GCC AGC TTG
Asn Phe Val Thr Leu Thr Asn Val Arg Asp Val Ile Ala Ser Leu

          729          738          747          756
GCG ATC ATG TTG TTT GTA TGC GGA GAG CGG CCA TCT -3'
Ala Ile Met Leu Phe Val Cys Gly Glu Arg Pro Ser
```

Fig. 11.b': Nucleotide sequence and derived amino acid sequence of rMLB

```
            9           18           27           36           45
5' GAT GAT GTT ACC TGC AGT GCT TCG GAA CCT ACG GTG CGG ATT GTG
   Asp Asp Val Thr Cys Ser Ala Ser Glu Pro Thr Val Arg Ile Val

           54           63           72           81           90
   GGT CGA AAT GGC ATG TGC GTG GAC GTC CGA GAT GAC GAT TTC CGC
   Gly Arg Asn Gly Met Cys Val Asp Val Arg Asp Asp Asp Phe Arg

           99          108          117          126          135
   GAT GGA AAT CAG ATA CAG TTG TGG CCC TCC AAG TCC AAC AAT GAT
   Asp Gly Asn Gln Ile Gln Leu Trp Pro Ser Lys Ser Asn Asn Asp

          144          153          162          171          180
   CCG AAT CAG TTG TGG ACG ATC AAA AGG GAT GGA ACC ATT CGA TCC
   Pro Asn Gln Leu Trp Thr Ile Lys Arg Asp Gly Thr Ile Arg Ser

          189          198          207          216          225
   AAT GGC AGC TGC TTG ACC ACG TAT GGC TAT ACT GCT GGC GTC TAT
   Asn Gly Ser Cys Leu Thr Thr Tyr Gly Tyr Thr Ala Gly Val Tyr
```

Fig. 11.b' continued:

```
                234         243         252         261         270
5'- GTG ATG ATC TTC GAC TGT AAT ACT GCT GTG CGG GAG GCC ACT CTT
    Val Met Ile Phe Asp Cys Asn Thr Ala Val Arg Glu Ala Thr Leu

                279         288         297         306         315
    TGG CAG ATA TGG GGC AAT GGG ACC ATC ATC AAT CCA AGA TCC AAT
    Trp Gln Ile Trp Gly Asn Gly Thr Ile Ile Asn Pro Arg Ser Asn

                324         333         342         351         360
    CTG GTT TTG GCA GCA TCA TCT GGA ATC AAA GGC ACT ACG CTT ACG
    Leu Val Leu Ala Ala Ser Ser Gly Ile Lys Gly Thr Thr Leu Thr

                369         378         387         396         405
    GTG CAA ACA CTG GAT TAC ACG TTG GGA CAG GGC TGG CTT GCC GGT
    Val Gln Thr Leu Asp Tyr Thr Leu Gly Gln Gly Trp Leu Ala Gly

                414         423         432         441         450
    AAT GAT ACC GCC CCA CGC GAG GTG ACC ATA TAT GGG TTC AGG GAC
    Asn Asp Thr Ala Pro Arg Glu Val Thr Ile Tyr Gly Phe Arg Asp

                459         468         477         486         495
    CTT TGC ATG GAA TCA AAT GGA GGG AGT GTG TGG GTG GAG ACG TGC
    Leu Cys Met Glu Ser Asn Gly Gly Ser Val Trp Val Glu Thr Cys

                504         513         522         531         540
    GTG AGT AGC CAA AAG AAC CAA AGA TGG GCT TTG TAC GGG GAT GGT
    Val Ser Ser Gln Lys Asn Gln Arg Trp Ala Leu Tyr Gly Asp Gly

                549         558         567         576         585
    TCT ATA CGC CCC AAA CAA AAC CAA GAC CAA TGC CTC ACC TGT GGG
    Ser Ile Arg Pro Lys Gln Asn Gln Asp Gln Cys Leu Thr Cys Gly

                594         603         612         621         630
    AGA GAC TCC GTT TCA ACA GTA ATC AAT ATA GTT AGC TGC AGC GCT
    Arg Asp Ser Val Ser Thr Val Ile Asn Ile Val Ser Cys Ser Ala

                639         648         657         666         675
    GGA TCG TCT GGG CAG CGA TGG GTG TTT ACC AAT GAA GGG GCC ATT
    Gly Ser Ser Gly Gln Arg Trp Val Phe Thr Asn Glu Gly Ala Ile

                684         693         702         711         720
    TTG AAT TTA AAG AAT GGG TTG GCC ATG GAT GTG GCG CAA GCA AAT
    Leu Asn Leu Lys Asn Gly Leu Ala Met Asp Val Ala Gln Ala Asn

                729         738         747         756         765
    CCA AAG CTC CGC CGA ATA ATC ATC TAT CCT GCC ACA GGA AAA CCA
    Pro Lys Leu Arg Arg Ile Ile Ile Tyr Pro Ala Thr Gly Lys Pro

                774         783     789
    AAT CAA ATG TGG CTT CCC GTG CCA -3'
    Asn Gln Met Trp Leu Pro Val Pro
```

Fig. 11.c': Nucleotide sequence and derived amino acid sequence of the rML propeptide

```
                  9           18
5'- TCC TCT GAG GTG CGC TAT TGG CCG
    Ser Ser Glu Val Arg Tyr Trp Pro

 27           36           45  48
CTG GTC ATA CGA CCC GTG ATA GCC -3'
Leu Val Ile Arg Pro Val Ile Ala
```

Fig. 11.d: Flanking regions of the ProML gene cassette in expression vector pT7ProML

```
   Nde I                                                    Bam HI
...TACAT ATG TAC                                CCA TGA TAA GGATCCTCTAG
         Met Tyr  ... ProML ...                 Pro *** ***
```

Fig. 11.e: Flanking regions of the IML gene cassette in expression vector pIML-02-P

```
   Stu I                                  Spe I     BseRI BamHI Xba I Sal I
...CAG GCC TAC                       CAC TAG TAA CTCCTCGGATCCTCTAGAGTCGACC...
   Gln Ala Tyr  ...IML...            His *** ***
```

Fig. 12: Recombinant production of rML

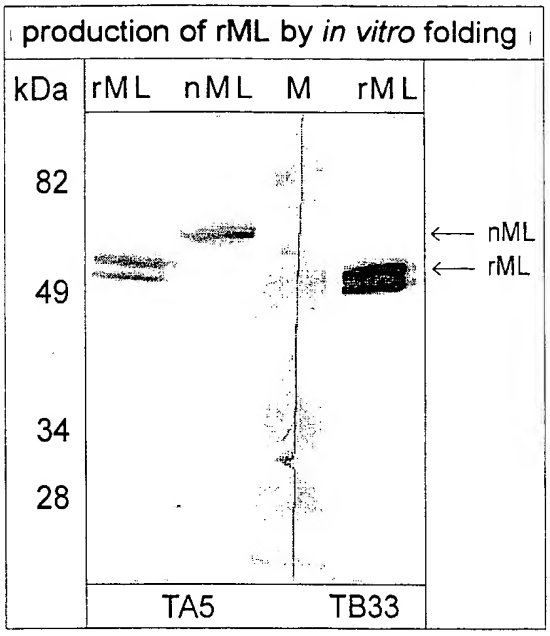


Fig. 13: Recombinant production of rIML (rMLΔ1α1β2γ)

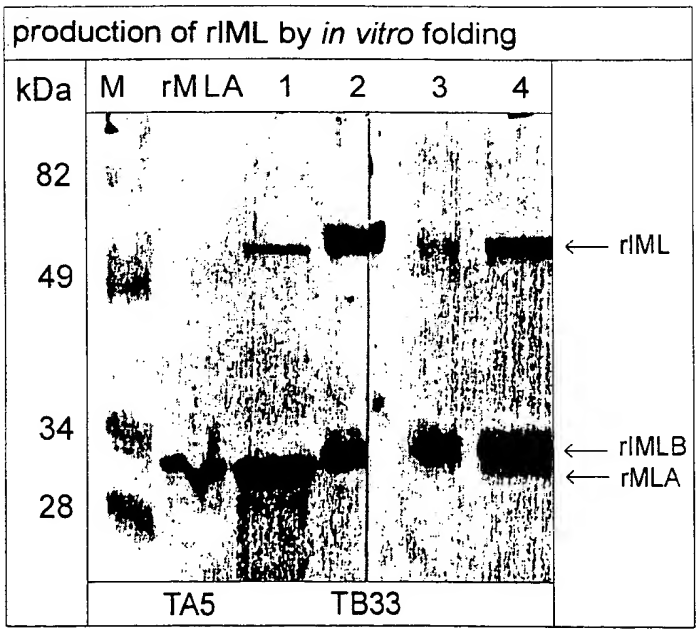


Fig. 14: Cytotoxicity of rIML with inactivated carbohydrate binding site as compared to rML (wild-type)

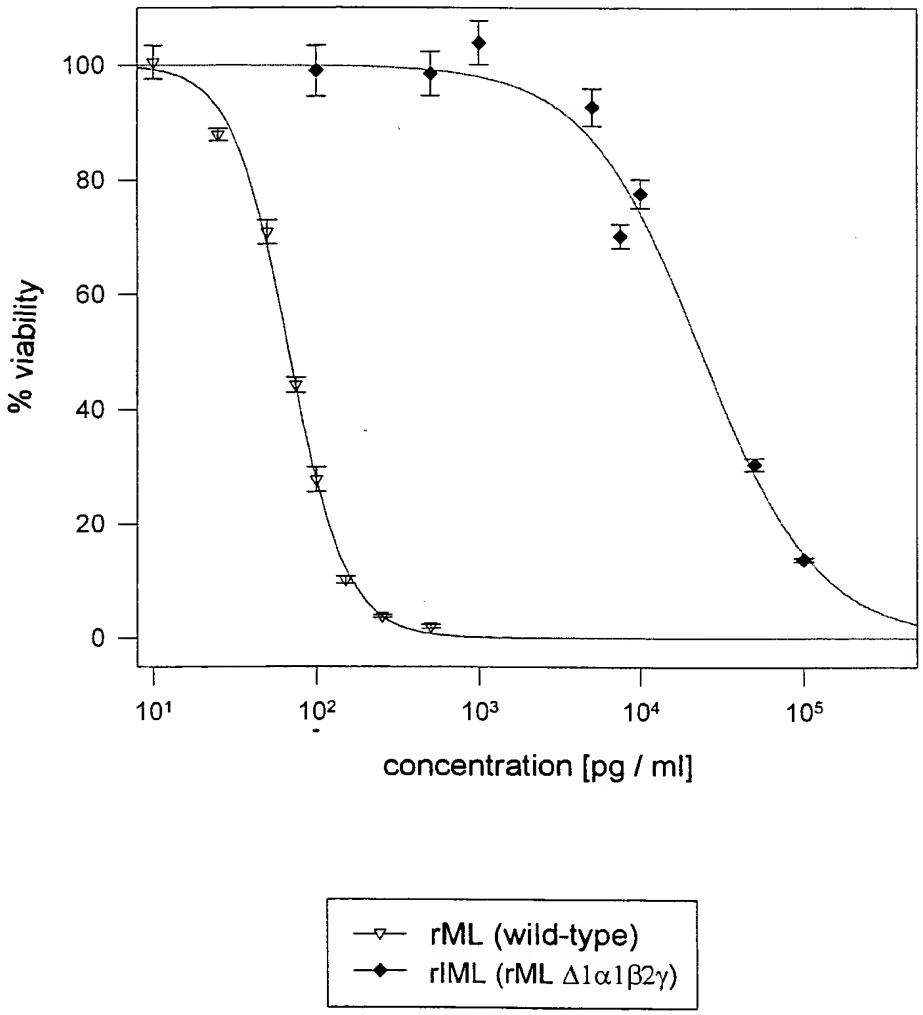


Fig. 15: Construction of a vector for the expression of an rMLB variant without carbohydrate affinity

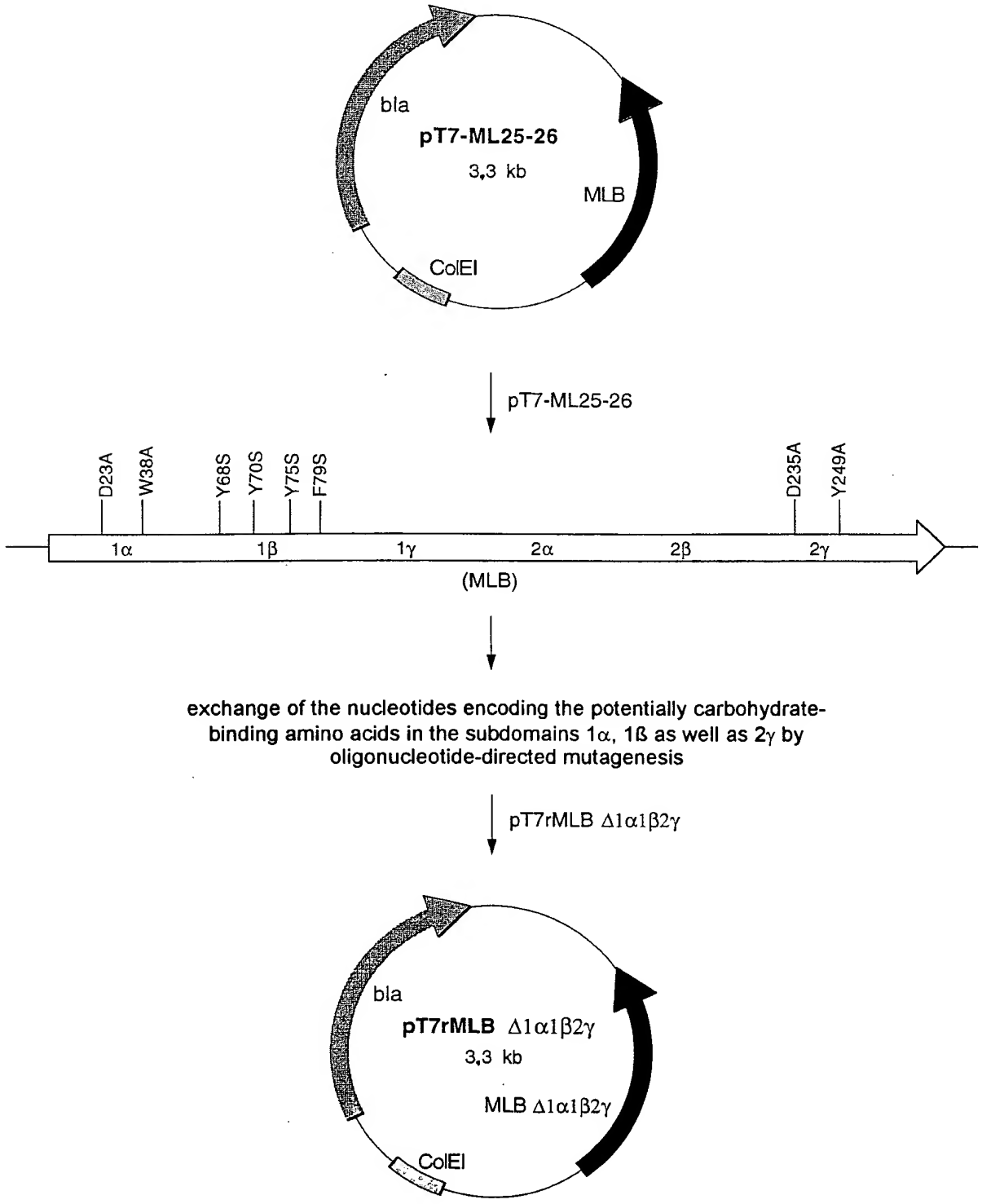


Fig. 16.1: Construction of a modular periplasmic expression system for the production of ITF toxins

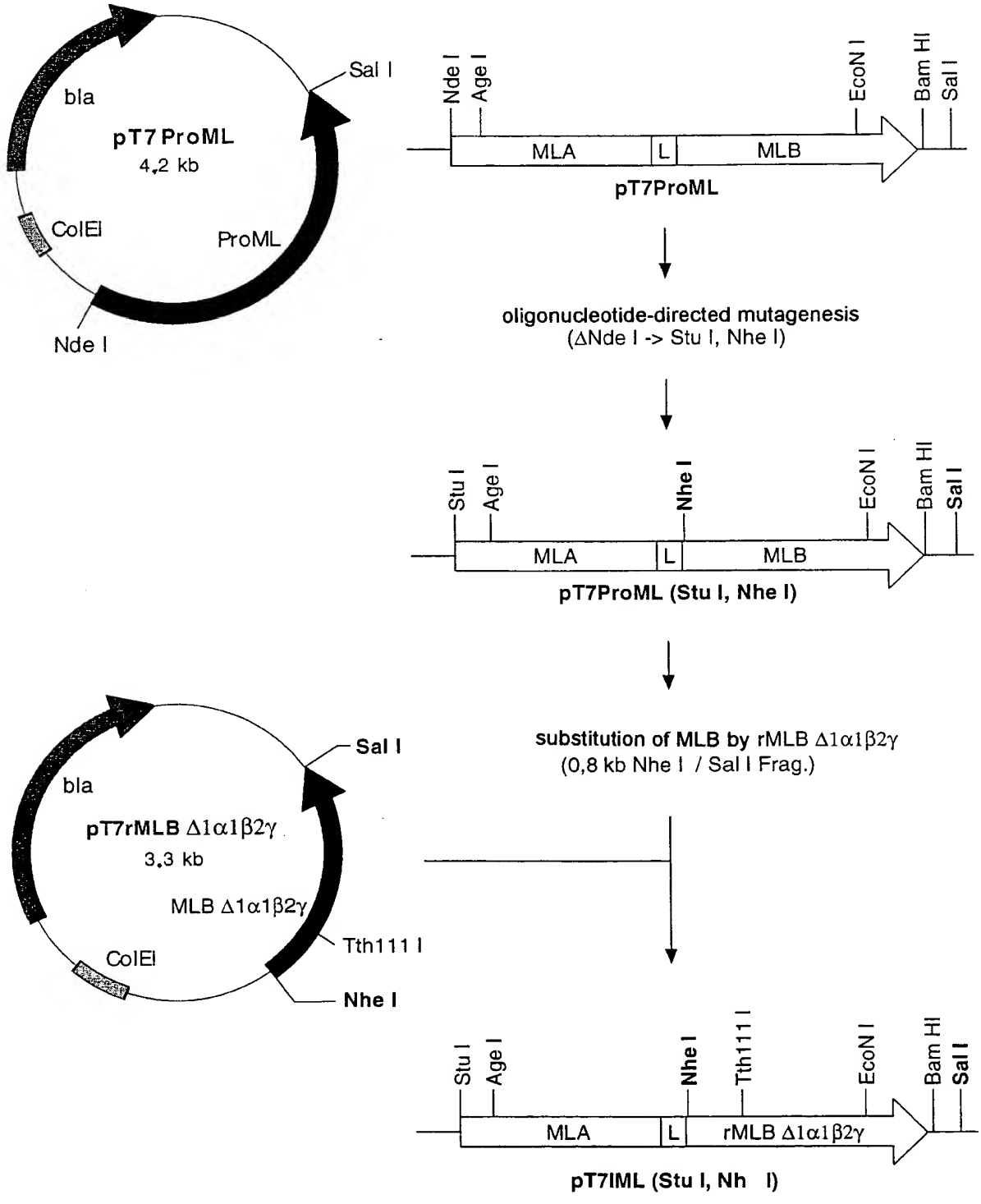


Fig. 16.2 (continued):

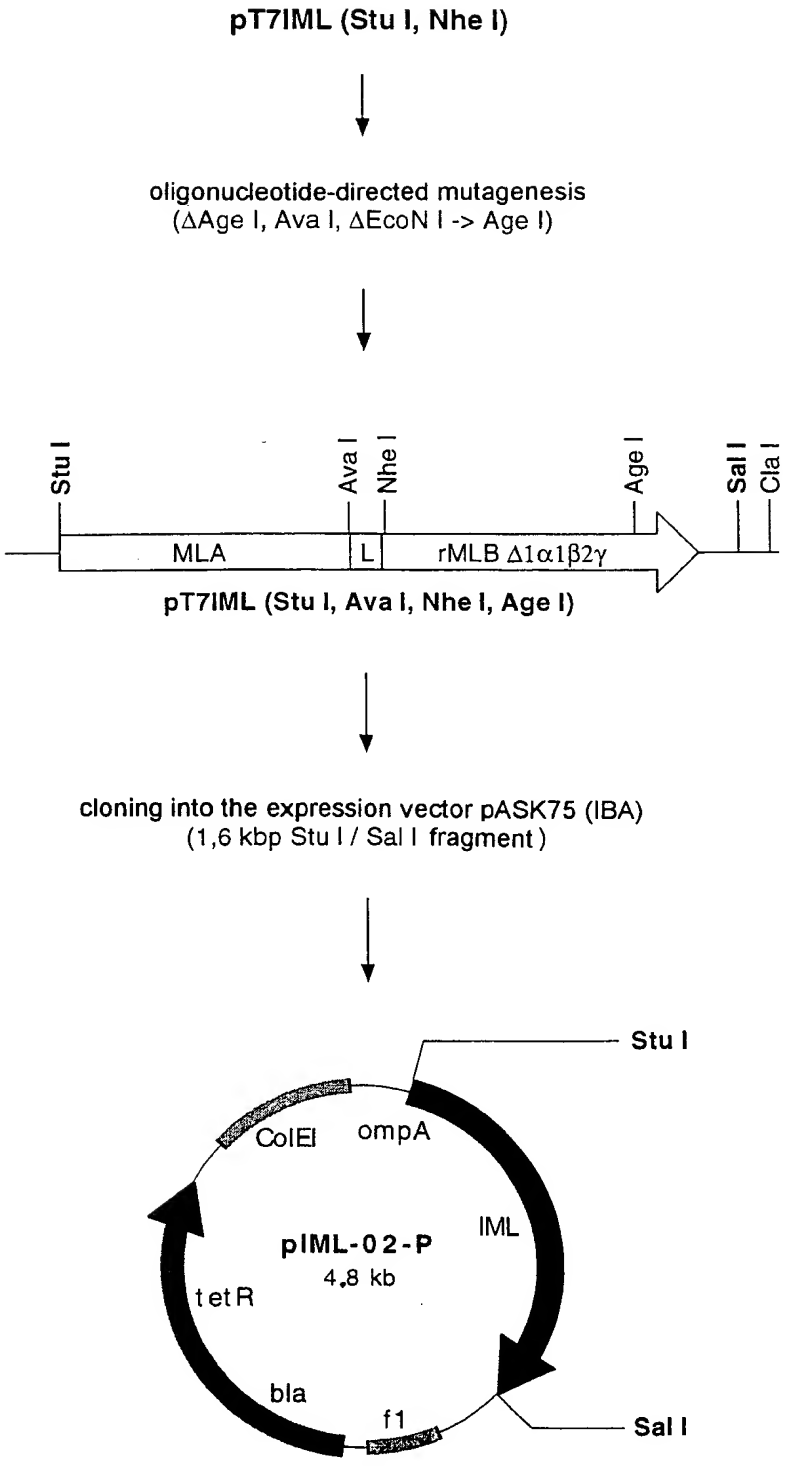


Fig. 16.3 (continued):

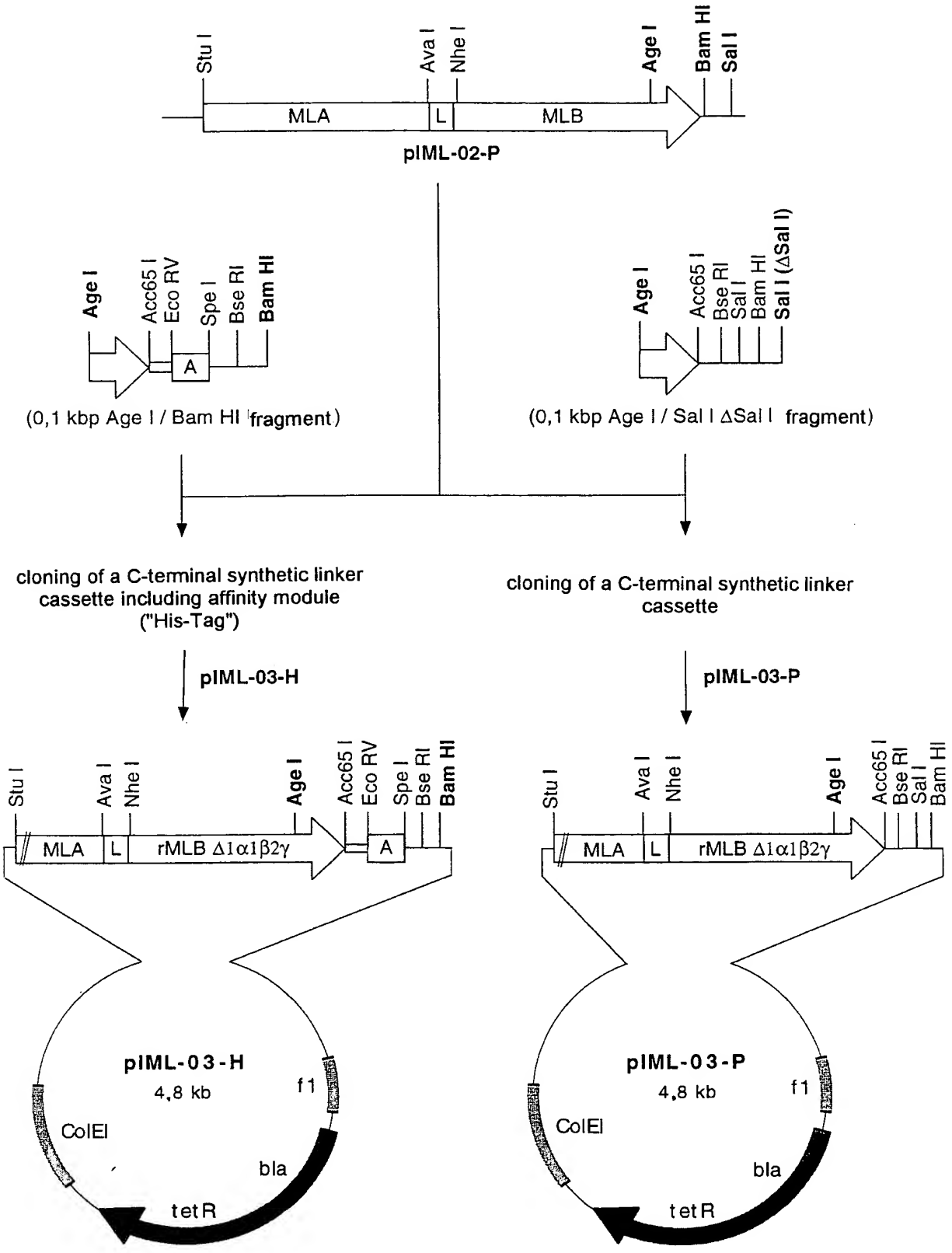
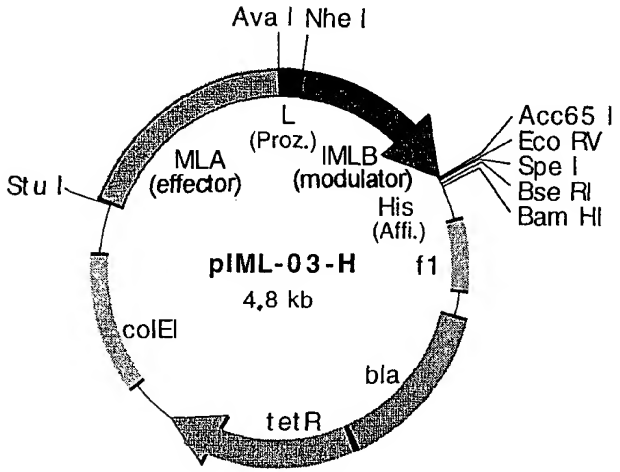
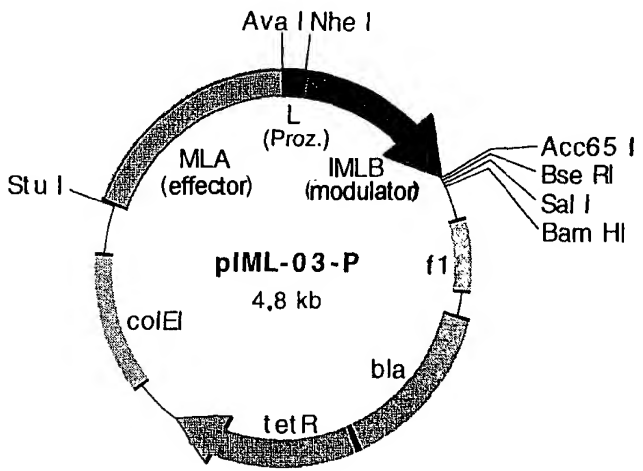
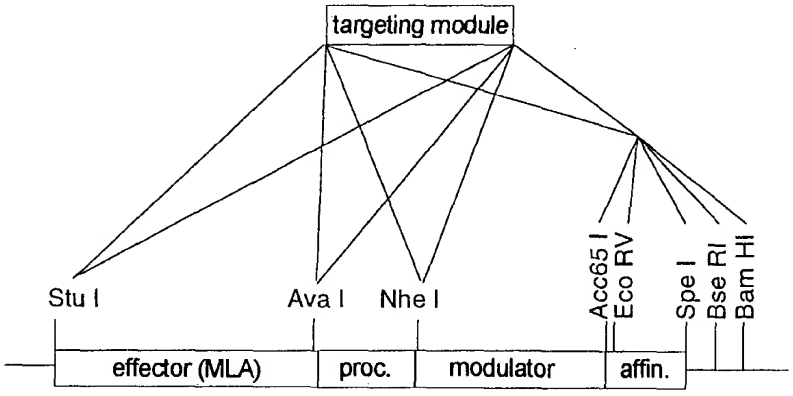


Fig. 17: Assembly of ITF toxins on the basis of vectors pIML-03-H or pIML-03-P with specific activity against target cells







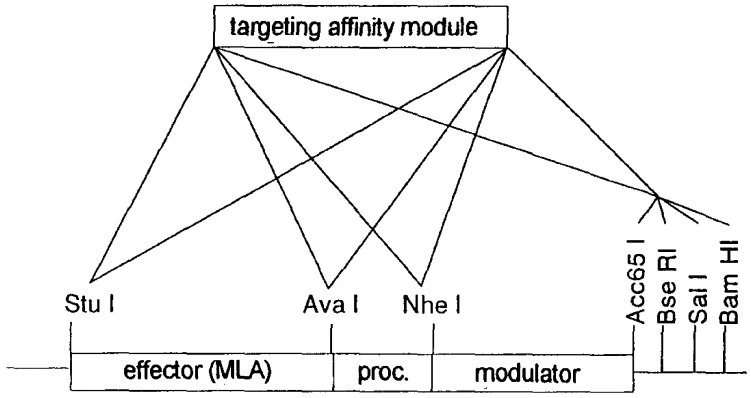


Fig. 18: Vector for the expression of an ITF toxin, specific for a P2-reactive neuritogenic T cell line

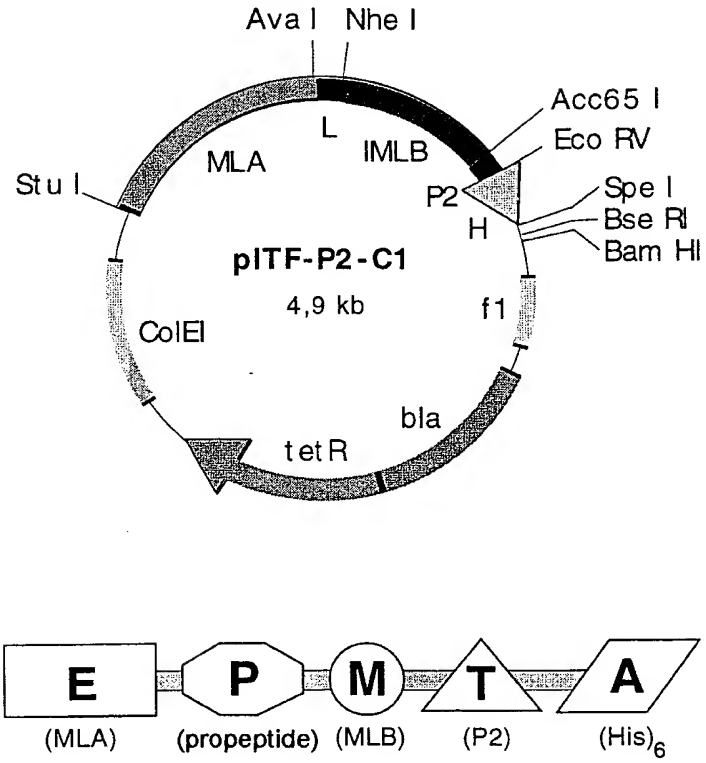


Fig. 19: Synthetic gene cassette encoding amino acids 53 to 78 of the P2 protein

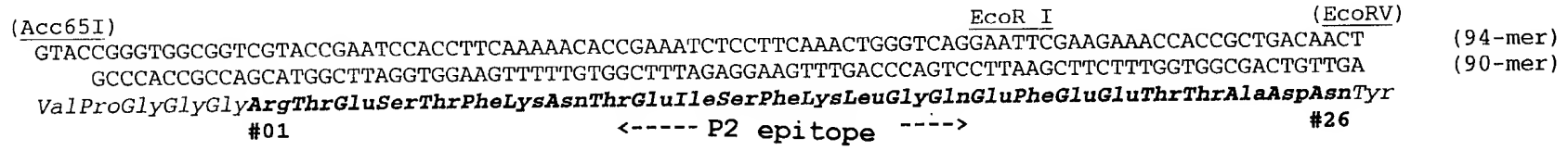


Fig. 20: Synthetic linker cassette for providing modularity at the 3' end of rMLB Δ1α1β2γ

```
Age I (ΔEcoNI)              Acc65I        BseR ISal I        BamH I SalI(ΔSalI)
CACCGGTAAACCGAACCAGATGTGGCTGCCGGTACCGTAGTAACGCTCCTCGTCGACCTAGTAAGGATCCC          69 mer
 TGGCCATTTGGCTTGGTCTACACCGACGGCCATGGCATCATTGCGAGGAGCAGCTGGATCATTCCTAGGGAGCT      69 mer
 ThrGlyLysProAsnGlnMetTrpLeuProValPro******
            <----  rMLB Δ1α1β2γ #522
```


Fig. 21: Synthetic linker cassette for providing modularity at the 3' end of rMLB Δ1α1β2γ with affinity module ("His-Tag")

```
Age  I(ΔEcoNI)              Acc65I        EcoR V                Spe I   BseR IBamHI
 CCGGTAAACCGAACCAGATGTGGCTGCCGGTACCGGGTGGTGGATATCATCACCACCATCACCACTAGTAACTCCTCG      78 mer
     ATTTGGCTTGGTCTACACCGACGGCCATGGCCCACCACCTATAGTAGTGGTGGTAGTGGTGATCATTGAGGAGCCTAG  78 mer
ThrGlyLysProAsnGlnMetTrpLeuProValProGlyGlyGlyTyrHisHisHisHisHisHis******
       <--  rMLB Δ1α1β2γ  -->  #552;  adapter  ;<--  His-Tag  -->
```

Fig. 22: Mutagenic oligonucleotides for the inactivation of carbohydrate binding sites in rMLB

1α domain:

1α$_1$ (D23A): 5'- CATGTGCGTGGCCGTCCGAGATGACG -3' (26-mer)
1α$_2$ (W38A): 5'- CAGATACAGTTGGCGCCCTCCAAGTCC -3' (27-mer)

1β domain:

1β (Y68S, Y70S, Y75S, F79S): 5'- GCTGCTTGACCACGTCTGGCTC...
...TACTGCTGGCGTCTCTGTGATGATCTCCGACTGTAATAC -3' (61-mer)

2γ domain:

2γ$_1$ (D235A): 5'- GGGTTGGCCATGGCTGTGGCGCAAGC -3' (26-mer)
2γ$_2$ (Y249A): 5'- CGAATAATCATCGCTCCTGCCACAGG -3' (26-mer)

pT7 selection primer:

pT7 Eco RV -> Ssp I: 5'- CTTCCTTTTTCAATATTATTGAAGCATTTATCAGG -3' (35-mer)
pT7 Ssp I -> Eco RV: 5'- CTTCCTTTTTCGATATCATTGAAGCATTTATCAGG -3' (35-mer)

Fig. 23: Mutagenic oligonucleotides for the construction of modular ITF gene cassettes

pT7 ΔNde I -> Stu I:
5'- CTTTAAGAAGGAGATATACAGGCCTACGAGAGGCTAAGAC -3' (40-mer)

pT7 Nhe I:
5'- GTTACCTGCAGTGCTAGCGAACCTACGGTGCGG -3' (33-mer)

pT7 ΔAge I:
5'- CCCACCAGACCACCGGCGAAGAATATTTCCGG -3' (32-mer)

pT7 Ava I:
5'- GTTTGTATGCGGAGAGCGTCCCTCGAGCTCTGAGGTGCGC -3' (40-mer)

pT7 IML ΔNde I -> Age I:
5'- CCGAATAATCATCGCTCCGGCCACCGGTAAACCAAATCAAATG -3' (43-mer)

Fig. 24: Purification of ITF-P2-C1 on Ni-NTA sepharose under denaturing conditions

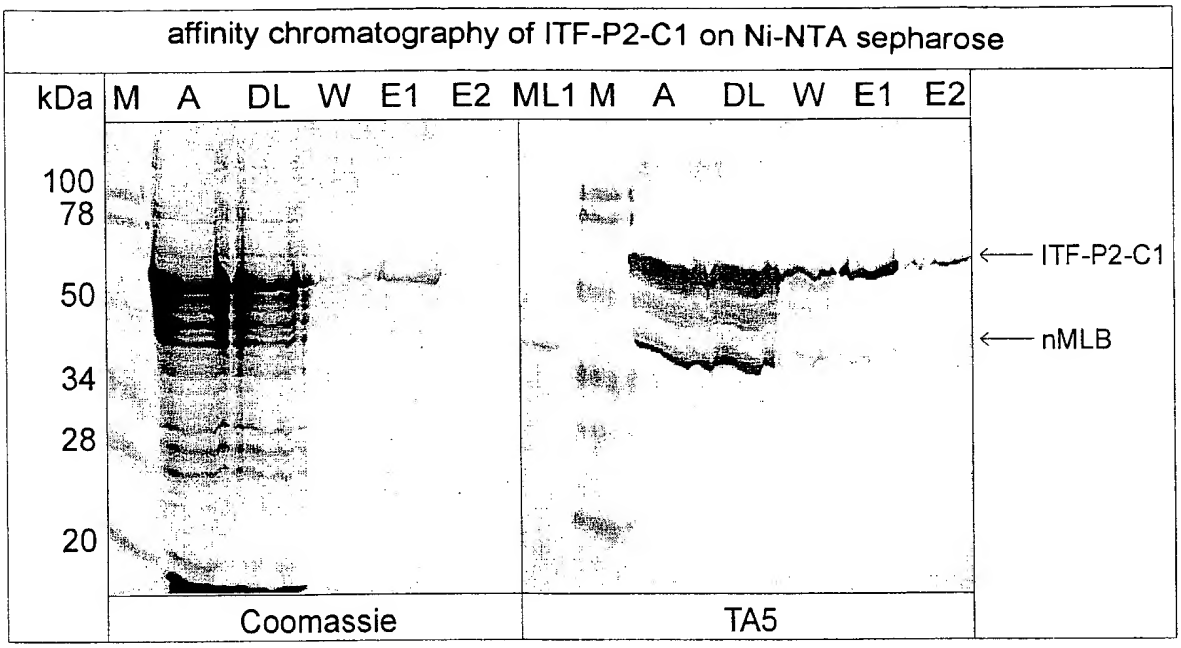


Fig. 25: Purification of ITF-P2-C1 on Ni-NTA sepharose under physiological conditions

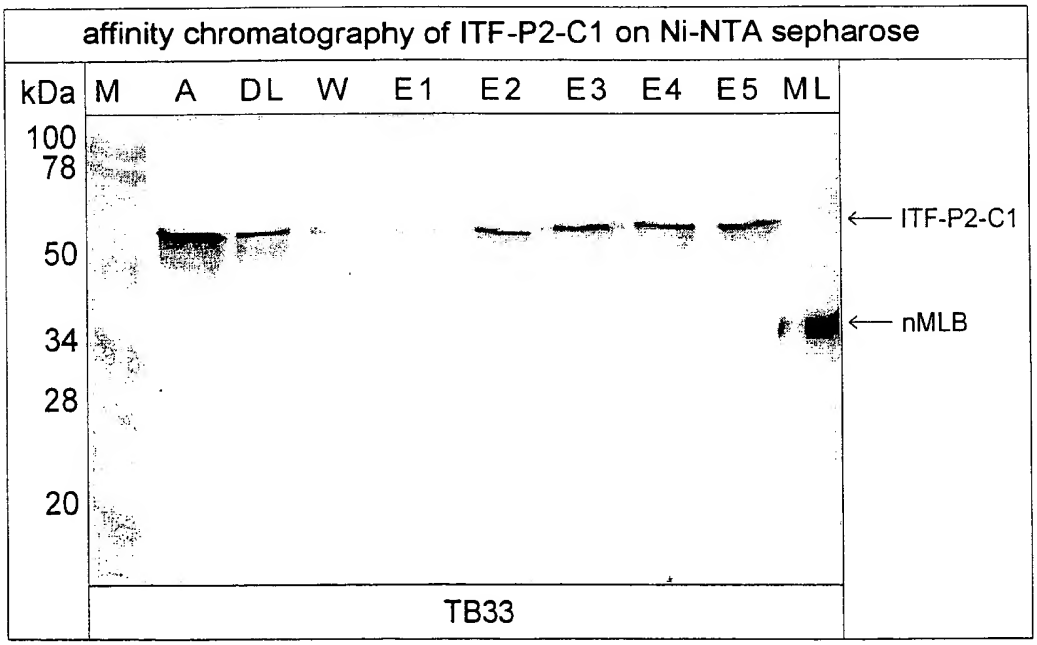


Fig. 26: Processing of pITF-P2-C1 during production in *E. coli*

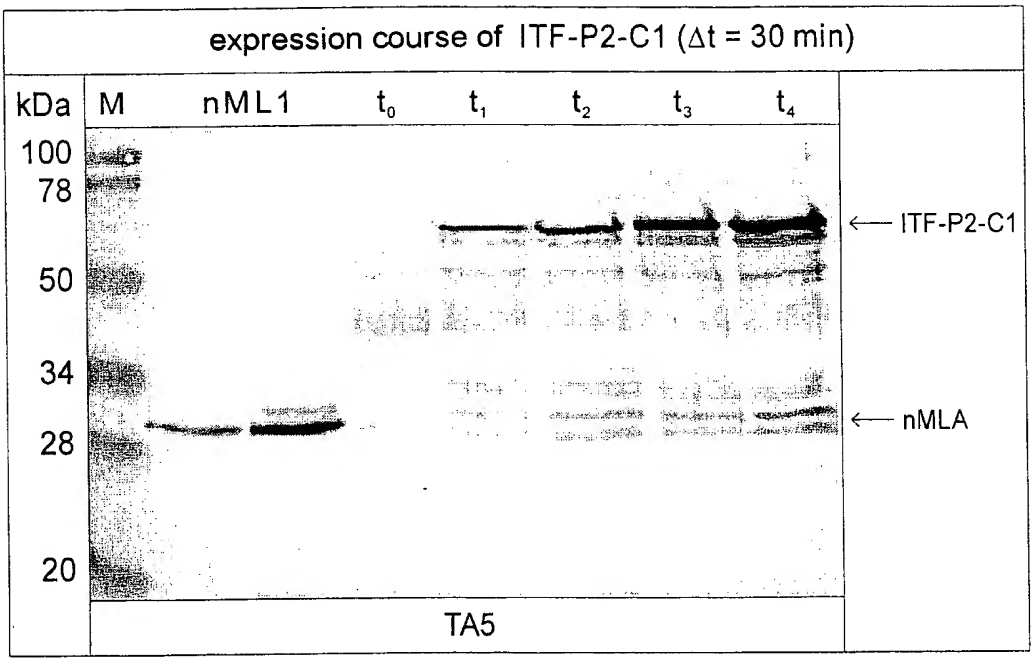


Fig. 27: Production of ITF by *in vitro* folding

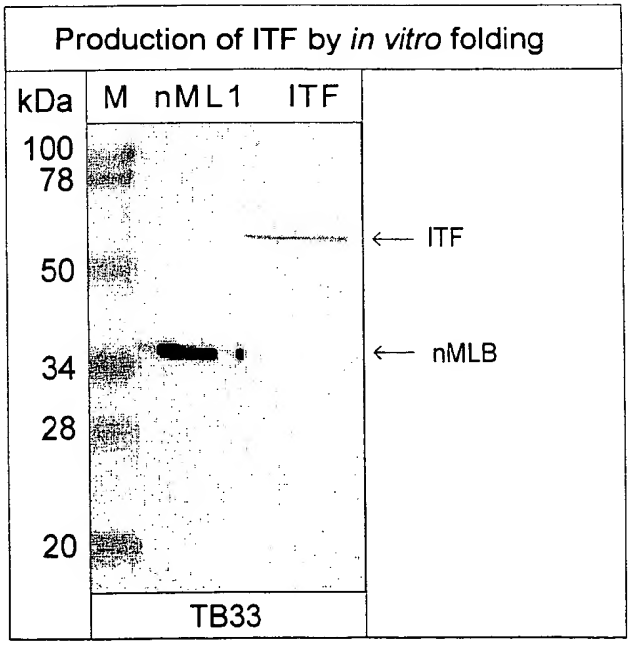
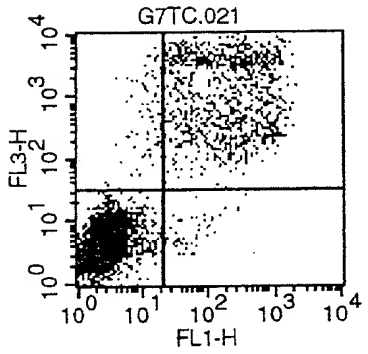
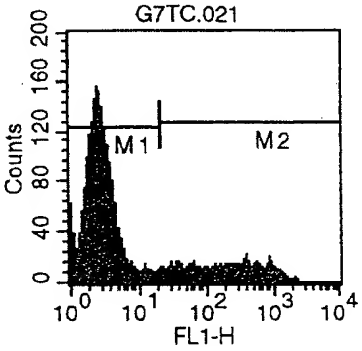


Fig. 28: FACS analysis of P2-specific T cells after incubation with ITF-P2-C1 for 2 hours

28.a

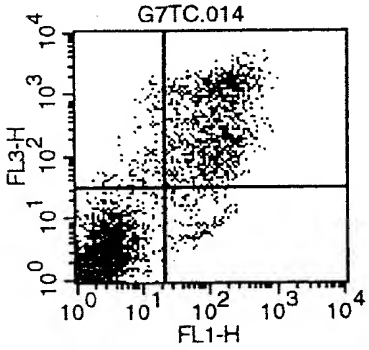


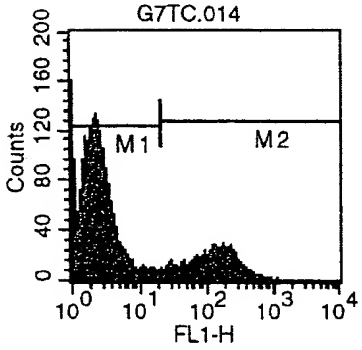


File: G7TC.021
Sample ID: T2 PUFFER 100UL
Gate: G1
Total Events: 5455
Y Parameter: FL3-H (Log)

Quad	Events	% Gated	% Total
UL	88	1.75	1.61
UR	1070	21.30	19.62
LL	3817	75.99	69.97
LR	48	0.96	0.88

28.b

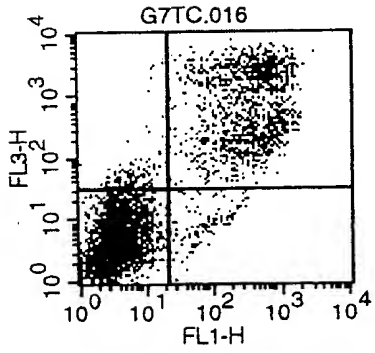


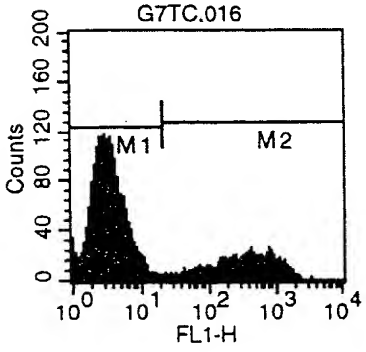


File: G7TC.014
Sample ID: T2 ITFCO1 50UL
Gate: G1
Total Events: 6111
Y Parameter: FL3-H (Log)

Quad	Events	% Gated	% Total
UL	140	2.78	2.29
UR	1121	22.26	18.34
LL	3643	72.35	59.61
LR	131	2.60	2.14

28.c



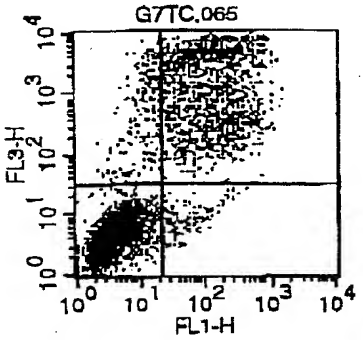


File: G7TC.016
Sample ID: T2 ITFCO1 100UL
Gate: G1
Total Events: 5569
Y Parameter: FL3-H (Log)

Quad	Ev nts	% Gated	% Total
UL	143	2.83	2.57
UR	1257	24.91	22.57
LL	3529	69.94	63.37
LR	117	2.32	2.10

Fig. 29: FACS analysis of P2-specific T cells after incubation with ITF-P2-C1 for 24 hours

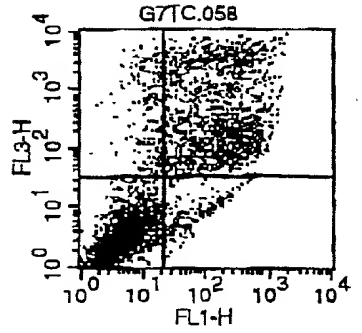
29.a.



File: G7TC.065
Sample ID: T24 PUFFER 100UL
Gate: G1
Total Events: 5662
Y Parameter: FL3-H (Log)

Quad	Events	% Gated	% Total
UL	209	4.16	3.69
UR	1467	29.22	25.91
LL	3207	63.88	56.64
LR	137	2.73	2.42

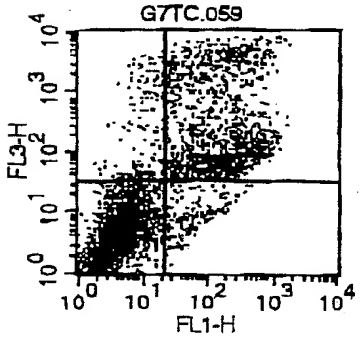
29.b.



File: G7TC.058
Sample ID: T24 ITFC01 50UL
Gate: G1
Total Events: 6043
Y Parameter: FL3-H (Log)

Quad	Events	% Gated	% Total
UL	212	4.20	3.51
UR	1620	32.13	26.81
LL	3018	59.86	49.94
LR	192	3.81	3.18

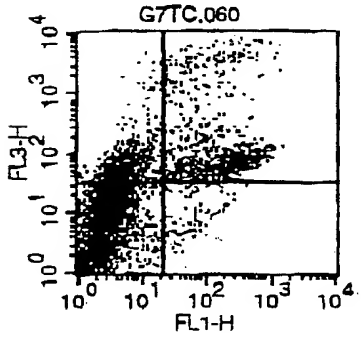
29.c.



File: G7TC.059
Sample ID: T24 ITFC01 75UL
Gate: G1
Total Events: 5780
Y Parameter: FL3-H (Log)

Quad	Events	% Gated	% Total
UL	207	4.10	3.58
UR	1449	28.72	25.07
LL	3209	63.59	55.52
LR	181	3.59	3.13

29.d.



File: G7TC.060
Sample ID: T24 ITFC01 100UL
Gate: G1
Total Events: 5627
Y Parameter: FL3-H (Log)

Quad	Events	% Gated	% Total
UL	834	16.57	14.82
UR	896	17.80	15.92
LL	3188	63.33	56.66
LR	116	2.30	2.06